INVESTIGATION OF THE ALTERATIONS IN MOTOR UNITS IN NEUROLOGIC DISORDERS BY SCANNING ELECTROMYOGRAPHY

by

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ABSTRACT

INVESTIGATION OF THE ALTERATIONS IN MOTOR UNITS IN NEUROLOGIC DISORDERS BY SCANNING ELECTROMYOGRAPHY

In this study, the alterations in the length of cross-sections of MU and the changes in maximum amplitude of MUAPs in each MU in patients with JME were investigated.

An experimental setup of scanning EMG was built and 3-D cross sectional maps of the MUs were plotted in order to measure the length of cross-sections and to find the maximum amplitude of each MU. Three subject groups comprising nine patients with juvenile myoclonic epilepsy as JME group, ten healthy volunteers as normal control (NC) group and three patients with spinal muscular atrophy as SMA group were included. The age of the subjects ranged between 22 and 46. Five to eight measurements were performed from the biceps brachialis muscles of each subject. Data including 113 measurements in total acquired from with these measurements were stored in a computer and then were used to construct 3-D maps of MU territories.

All three groups were compared in pairs by using 113 measurements with Student's t-test. JME groups were found similar to SMA group in terms of both parameters. The difference between JME and NC groups was found as extremely significant. Since the increase in both parameters due to the reinnervation occurs in SMA group, significant difference is expected between SMA and NC group. These results were confirmed with Tukey's HSD test by comparing three groups simultaneously. Three groups were also compared using the individual means of parameters with a non-parametric test such as Mann-Whitney test. A significant difference which is also confirmed again with Tukey's HSD test was found between the JME and NC groups.

In conclusion, since no neurogenic evidence was found in JME patients in conventional EMG previously higher length of cross-sections can be considered as structural.

Keywords: Scanning EMG, Juvenile Myoclonic Epilepsy, Motor Unit Territory, Electrophysiological Cross-section.

ÖZET

NÖROLOJİK BOZUKLUKLARDA MOTOR ÜNİTELERDEKİ DEĞİŞİKLİKLERİN TARAMALI ELEKTROMİYOGRAFİ İLE ARAŞTIRILMASI

Bu çalışmada, jüvenil miyoklonik epilepsili hastaların motor ünitelerinin kesit uzunluklarındaki ve motor ünite aksiyon potansiyellerinin maksimum genliklerindeki değişimler araştırılmıştır.

Ölçülen her bir motor ünitenin kesit uzunluğunu ölçmek amacıyla, ölçülen motor ünitelerin üç boyutlu kesit alanı haritalarını oluşturmak ve maksimum genliklerini bulmak üzere bir deneysel taramalı EMG düzeneği kurulmuştur. Dokuz jüvenil myoklonik epilepsili hastadan oluşan JME grubu, on sağlıklı gönüllülerden oluşan normal kontrol (NK) grubu ve üç spinal müsküler atrofi hastasından oluşan SMA grubu olmak üzere üç denek grubu oluşturuldu. Deneklerin yaşları 22 ila 46 arasındaydı. Her bir denekte, biseps brakialis kasından beş ile sekiz ölçüm uygulandı. Toplam olarak 113 ölçümden oluşan veriler bilgisayarda depolanarak motor ünite alanlarının haritalarını oluşturmak üzere kullanıldı.

Her üç grup ikişerli olarak 113 ölçüm kullanılarak Student's t-testi ile karşılaştırıldı. JME grupları her iki parametre yönünden SMA grubuna benzer bulundu. JME ve NK grupları arasındaki fark oldukça anlamlı bulundu. SMA grubundaki reinnervasyon nedeniyle her iki parametrede artışa bağlı olarak, SMA ve NK grupları arasındaki anlamlı bir fark beklenmekteydi. Bu sonuçlar her üç grubu Tukey HSD testiyle karşılaştırılarak doğrulandı. Parametrelerin her bir bireye ait ortalamaları parametrik olmayan Mann-Whitney testi ile karşılaştırıldı. JME ve NK grupları arasında Tukey HSD testiyle de doğrulanan anlamlı farklar bulundu.

Sonuç olarak, JME hastalarında daha önceki geleneksel EMG incelemelerinde her hangi bir nörojenik bulgu olmadığından, MU kesit uzunluklarının yüksek bulunması yapısal olarak kabul edilebilir.

Anahtar Kelimeler: Taramalı EMG, Jüvenil Miyoklonik Epilepsi, Motor Ünite Alanı, Elektrofizyolojik Kesit Alanı.

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C_{body}	Capacitance between body and earth
$C_{c1,}$ C_{c2}	Parasitic capacitances between power line and input cables
C_{ground}	Parasitic capacitance of the power supply between power line and isolated ground
C_{iso}	Parasitic capacitance of the isolation barrier
C_{power}	Parasitic capacitance between body and power line
N	Number of M-waves (CMAP)
V_c	Common mode input voltage
V_d	Differential input
V_{iso}	Isolation mode voltage
x	Individual M-waves
$Z_{electrode-1}$, $Z_{electrode-2}$	Electrode contact impedances
$Z_{input-1}$, $Z_{input-2}$	Amplifier input impedances
Z_{ref}	Patient reference contact impedance
σ^2	Variance

LIST OF ABBREVIATIONS

A/D	Analog-to-digital
A/T	Amplitude per turn
ACh	Acetyl choline
AChE	Acetylcholinesterase
ADC	Analog-to-digital converter
ADM	Abductor Digiti Minimi
ADQ	Abductor Digiti Quinti
ADQP	Abductor Digiti Quinti Pedis muscle
AED	Antiepileptic drugs
AEP	Auditory Evoked Potential
Ag	Silver
AgCl	Silver Chloride
AHB	Abductor Hallucis Brevis muscle
AIDP	Acute inflammatory demyelinating polyneuropathy
AIPE	Amplitude of the IP envelope
ALS	Amyotrophic lateral sclerosis
anti GM_1	Antiganglioside antibodies
AP	Action Potentials
APB	Abductor Pollicis Brevis
ATP	Adenosine tri phosphate
ATPase	Adenosine tri phosphatase
BB	Biceps brachialis
C1-C8	Cervical levels
Ca^{++}	Calcium ions
Ca-ATPase	Calcium-Adenosine tri phosphatase
CACNB4	Gene encoding calcium channel β subunit
CAE	Childhood absence epilepsy
CIDP	Chronic Inflammatory Demyelinating Polyneuropathy
CK	Creatinin Kinase
CMAP	Compound muscle action potential
CMRR	Common Mode Rejection Ratio
CMT	Charcot-Marie-Tooth

CNE	Concentric Needle Electrode
CNEMG	Concentric Needle Electromyography
CoA	Coenzyme A
CPK	Creatinin Phosphokinase
CPT	Carnitine palmitoyltransferase
CRD	Complex Repetitive Discharge
CRT	Cathode Ray Tube
CTS	Carpal tunnel syndrome
D/A	Digital-to-analog
DAC	Digital-to-analog converter
DAQ	Data Acquisition
DC	Direct Current
DM	Dermatomyositis
EDB	Extensor Digitorum Brevis muscle
EDC	Extensor digitorum communis
EDX	Electrodiagnosis
EEG	Electroencephalography
EHL	Extensor hallicus longus
EIP	Extensor indicis poroprius
EMG	Electromyography
EPP	End-plate potential
EQUIP	Expert Quantitative Interference Pattern Analysis
FD	Fiber density
FDI	First dorsal interosseous
FF	Fast-twitch fatigable
FIFO	First-In First-Out
FP	Fibrillation Potential
FR	Fast-twitch fatigue resistant
FSHD	Facioscapulohumeral Muscular Dystrophy
GABRA1	Gene encoding the alpha1 subunit of the gamma-amino-butyric acid receptor subtype A
$GABA_A$	Gamma-amino-butyric acid receptor subtype A
GBS	Guillain-Barré syndrome
GTCS	Generalized tonic clonic seizures

HF	High Frequency
HLA	Human Leukocyte Antigen
HMSN	Hereditary motor and sensory neuropathy
HSD	Honestly Significant Difference
HUA	Humeral-Ulnar aponeurosis
I/O	Input/Output
IBM	Inclusion body myositis
IGE	Idiopathic generalized epilepsy
IP	Interference Pattern
IPA	Interference Pattern Analysis
IPI	Interpotential interval
JAE	Juvenile absence epilepsy
JME	Juvenile myoclonic epilepsy
L1-L5	Lomber levels
LA	Linear Actuator
LAN	Local Area Network
LCD	Liquid Crystal Display
LEMS	Lambert-Eaton myasthenic syndrome
LMN	Lower Motor Neuron
MA	Mean Amplitude
MD	Muscular Dystrophy
MEPP	Miniature end-plate potential
MFAP	Muscle Fiber Action Potential
MG	Myastenia Gravis
MMNCB	Multifocal Motor Neuropathy with Conduction Block
MN	Motor Neuron
MND	Motor Neuron Disease
MNE	Monopolar Needle Electrode
MPS	Multiple point stimulation
MU	Motor Unit
MUAP	Motor Unit Action Potential
MUNE	Motor Unit Number Estimate
MUP	Motor Unit Potential
MUT	Motor Unit Territory

MVC	Maximum Voluntary Contraction
NC	Normal Control
NCS	Nerve Conduction Study
NCV	Nerve Conduction Velocity
NMD	Neuromuscular Disease
NMJ	Neuromuscular Junction
NSS	Number of Small Segment
PB	Peroneal Brevis
PCI	Peripheral Component Interconnect
PD	Parkinson's Disease
PIN	Posterior interosseous neuropathy
PL	Peroneal Longus
PM	Polymyositis
PPS	Post-polio syndrome
PRA	Peak ratio analysis
PSA	Power Spectrum Analysis
PSW	Positive Sharp Wave
Psw	Polyspike waves
PTH	Parathyroid hormone
RNS	Repetitive Nerve Stimulation
RS-232	Recommended Standard-232
RT	Rise Time
SD	Standard Deviation
SFAP	Single Fiber Action Potential
SFE	Single Fiber Electrode
SFEMG	Single Fiber Electromyography
SMA	Spinal Muscular Atrophy
SMUP	Single Motor Unit Potential
SNAP	Sensory nerve action potential
SNR	Signal-to-noise ratio
SR	Sarcoplasmic Reticulum
SR	Sampling Rate
s-SFEMG	Stimulation single-fiber EMG
STA	Spike-trigger averaging

S-Type	Slow-type
SW	Slow-Wave
T/S	Turn per second
TA	Tibialis anterior muscle
TAA	Turn/Amplitude analysis
TnC	Calcium binding troponin complex
TnI	Inhibitory subunit of troponin complex
TnT	Tropomyosin binding troponin complex
TTS	Tarsal tunnel syndrome
UCA	Upper Centile Amplitude
UMN	Upper Motor Neuron
USB	Universal Serial Bus
VEP	Visual Evoked Potential
VGCC	Voltage-gated calcium channels
v-SFEMG	Voluntary single-fiber EMG

1. INTRODUCTION

1.1 Background:

The ability to move in the surrounding environment is an essential characteristic of animals. The actuators of movement are the muscles whose contractions generate forces on the skeletal segments to which they are connected [1]. The musculoskeletal system is responsible of regulation of force output for precise movements, the maintenance of upright posture, locomotion and the performance of the gestures [2]. These tasks are controlled by electrical signals conveyed between the muscles and the peripheral and central nervous system and achieved by means of skeletal muscle contractions [3]. These contractions are initiated by the electrical activities of the muıscle fibers and are achieved via the conversion of the chemical energy and electrical energy into mechanical energy in order to establish the motor activity [4].

In order to initiate the locomotor activity the motor programming takes place in the premotor cortex [2]. The excitatory impulses are conveyed from the central nervous system to a target muscle via the primary motor cortex, the corticospinal tract and α-motor neurons of the anterior horns of the spinal cord [4, 5, 6]. At the end, the nerve fiber of one α-motor neuron branches and each branch contacts a single and specific muscle fiber [5]. Neuromuscular junction ensures the connection of the branches of the motor neuron with these muscle fibers by forming a chemical synapse between the motor neuron and the muscle fiber [5, 7]. Since muscle tissue is made of excitable cells like nerves, the contraction is initiated by the excitation of the muscle fibers following the chemical transmission taking place in the Neuromuscular Junction [2, 4]. When the membrane of the muscle is excited an action potential propagating alone the membrane of this fiber is generated at the post-synaptic part of the fiber. This action potential propagates inside the muscle fibers through the T-tubules of the fiber and initiates the

excitation-contraction coupling process followed by the muscle contraction to generate the force necessary for the movements.

A motor neuron, the myelin sheath of the motor neuron, the neuromuscular junction between the motor neuron and the muscle fibers innervated by this neuron and all these muscle fibers constitute together the Motor Unit that is the basic structural and functional units of skeletal muscles [4, 5, 6, 8, 9]. The electrical activities of the muscle fibers belonging to a motor unit form the Motor Unit Action Potential (MUAP) [2].

The Electromyography (EMG) is an electrophysiological method to study the motor unit action potential. It is accepted as a useful method in the diagnosis of the neuromuscular diseases. EMG is used to detect and characterize disease processes affecting the motor units and to provide a guide to prognosis [10].

It has been shown that chromosome-6 is linked to Juvenile Myoclonic Epilepsy (JME). Because of the genetic involvement of chromosome 6 in JME and in spinal development, it is suggested that an anterior horn cell involvement may accompany to JME [11-13]. As a result an increase in the motor unit diameter is expected. A subclinical anterior horn cell involvement with different features could be observed among JME patients by using some methods based on EMG measurements such as concentric needle electrode and turn/amplitude analysis [11, 14, 15]. In previous studies using the Macro EMG and the Motor Unit Number Estimate (MUNE) methods, some evidences are found suggesting the presence of normal large motor units replaced normal small motor units, rather than the presence of reinnervated motor units [11, 15]. Nevertheless, since either the parameters that they measure are affected by reinnervation process as in case of Macro EMG [11] or they are based on estimation as in case of MUNE, these techniques mentioned in the preceding paragraphs cannot give an absolute result about the diameter of a motor unit [11]. Therefore, a method that gives information

about the temporal and spatial characteristics of a motor unit such as Scanning EMG is needed. Scanning EMG provides data about the anatomical structure of the Motor Units beside their electrical features [8, 16, 17]. Therefore, it is possible to measure the length of cross-section of the motor unit and to get absolute information about the territory of the motor unit [8]. Thus, scanning EMG can help to confirm the preponderance of large motor units in JME cases.

1.2 Objective:

The aim of this study is to reveal the structural alterations in motor units in skeletal muscles in patients with JME by means of scanning EMG method and to reveal the preponderance of large motor units in the muscle of JME patients.

1.3 Motivation:

The clumsiness of the JME patients in performing fine movements has been dedicated to the myoclonic jerks. On the other hand, it has been suggested that the clumsiness might originate from the structural changes in motor units such as the preponderance of large motor units due to the subclinical anterior horn cell involvement found from the previous studies. To reveal the clumsiness of these patients is whether due to the myoclonic jerks or due to the large motor units might be helpful to the clinicians in building therapy strategies to improve the ability of these patients in establishing the fine movements.

1.4 Experimental Design of the Study:

The experimental design of the study is based on comparing JME subjects with other control groups in terms of length of cross-sections and maximum amplitudes of the motor units (MUs) under the investigation. Thus, three subject groups were formed such JME, normal control (NC) and spinal muscular atrophy (SMA) groups. SMA groups were formed due to the reinnervation process that may cause increases in length of cross-sections and in maximum amplitudes of motor units. Therefore, it is expected that JME group will be similar to SMA group in terms of these variables provided that there is a preponderance of large MUs in JME subjects.

An experimental setup for scanning EMG method was designed and was achieved to gather data for this purpose.

1.5 Outline:

In Chapter 2, the anatomical structures, the physiological processes playing role in muscle contraction and the generation of electrical activity during the contraction are described.

In Chapter 3, the principles of electromyography, the structure and the components of the EMG instrument, the electrical activity detected by EMG, the special EMG techniques including Scanning EMG are explained.

Chapter 4 mentions to the neuromuscular disorders being under the scope of the electromyography are introduced.

Chapter 5 depicts the general characteristics of the juvenile myoclonic epilepsy.

In Chapter 6, the experimental setup of Scanning EMG used in this study, the measurement procedures, patient selection criteria and the statistical methods used in analyzing acquired data are described.

In Chapter 7, the results obtained from the statistical assessment of the acquired data are discussed.

This study is finally concluded in Chapter 8.

The figures and the schemas in this dissertation were plotted by using Microsoft® Office Visio® Professional 2003 application software.

2. ANATOMY AND PHYSIOLOGY OF THE SKELETAL MUSCLE

2.1 Introduction

The mobility of the body and the movements of many internal organs are established by converting the chemical energy into the mechanical by the muscles of diverse types. Beside these motions, the ability to communicate by speech, by writing or by gestures is ensured by the contractions of the muscles [7, 18].

There are three general types of muscles: (1) skeletal muscles used primarily for moving the bones; (2) smooth muscles surrounding the hollow organs such as stomach, bowels, blood vessels used for either propelling the luminal content of these organs via peristalsis or for regulating the internal flow by changing the diameter; (3) cardiac muscle being used for pumping the blood [7, 19].

Although the force-generating mechanisms are similar in all of these types of muscles, since they can be contracted voluntarily the skeletal muscles are different from other types [7]. Skeletal muscle has primarily four functions for the organism of human beings.

i. Production of Movement: Skeletal Muscles are responsible for all locomotion to enable the organism to respond quickly to changes in external environment. Furthermore, some skeletal muscle play role in changing the direction of the eyeballs to help the vision. Gestures are established by the contraction of the facial muscles. The speech is achieved also by the contraction of some muscle including those governing the movements of the tongue and those of lips.

ii. Maintaining Posture: Some skeletal muscles are responsible of maintaining the equilibrium of the body either in erect or seated position in the presence of the gravity
iii. Stabilizing Joints: Skeletal muscles can help stabilize the joints such as shoulder and knee with poor reinforcement and that do not possess complementary surface
iv. Generating Heat: As they contract the muscles produce heat. Since skeletal muscles account 40% of body mass, this heat has a vital importance in maintaining normal body temperature [18].

2.2 Anatomical Structure of the Skeletal Muscle

2.2.1 Gross Anatomy of a Skeletal Muscle

The muscles are made up of hundreds to thousands muscles fibers. The muscle is grouped into bundles called fascicles that are surrounded by connective tissue [7, 18] [20]. Muscles are joined to bones by bundles of collagen fibers known as tendons [7, 18] [20]. When muscle fibers contract they pull the tendons to transmit the force to the bone to be moved [18]. The organization of skeletal muscle that is attached to bones by tendons is represented in Figure 2.1

2.2.2 Microscopic Anatomy of a Skeletal Muscle

The skeletal muscle is made of long cylindrical cells called muscle fibers. They are multinucleated cells with diameters ranging from 10 to 100 μm and with lengths extending up to 20 cm. These nuclei are beneath the cell membrane of the muscle fiber which is called sarcolemma. Each muscle fiber is surrounded by sarcolemma. It contains T-tubules playing role in the propagation of the muscle action potential. The cytoplasm of

the muscle fiber is called sarcoplasma. It contains large amounts of stored glycogen and myoglobin that does not exist in other cell types. Myoglobin is an oxygen-binding protein and stores oxygen in muscle fiber [7, 18].

When the muscle fiber of skeletal muscle is observed through a light microscope, a series of light and dark bands are perpendicular to long axis of the fiber are seen. Due to this characteristic the skeletal muscle is also known as striated muscle bridges [7].

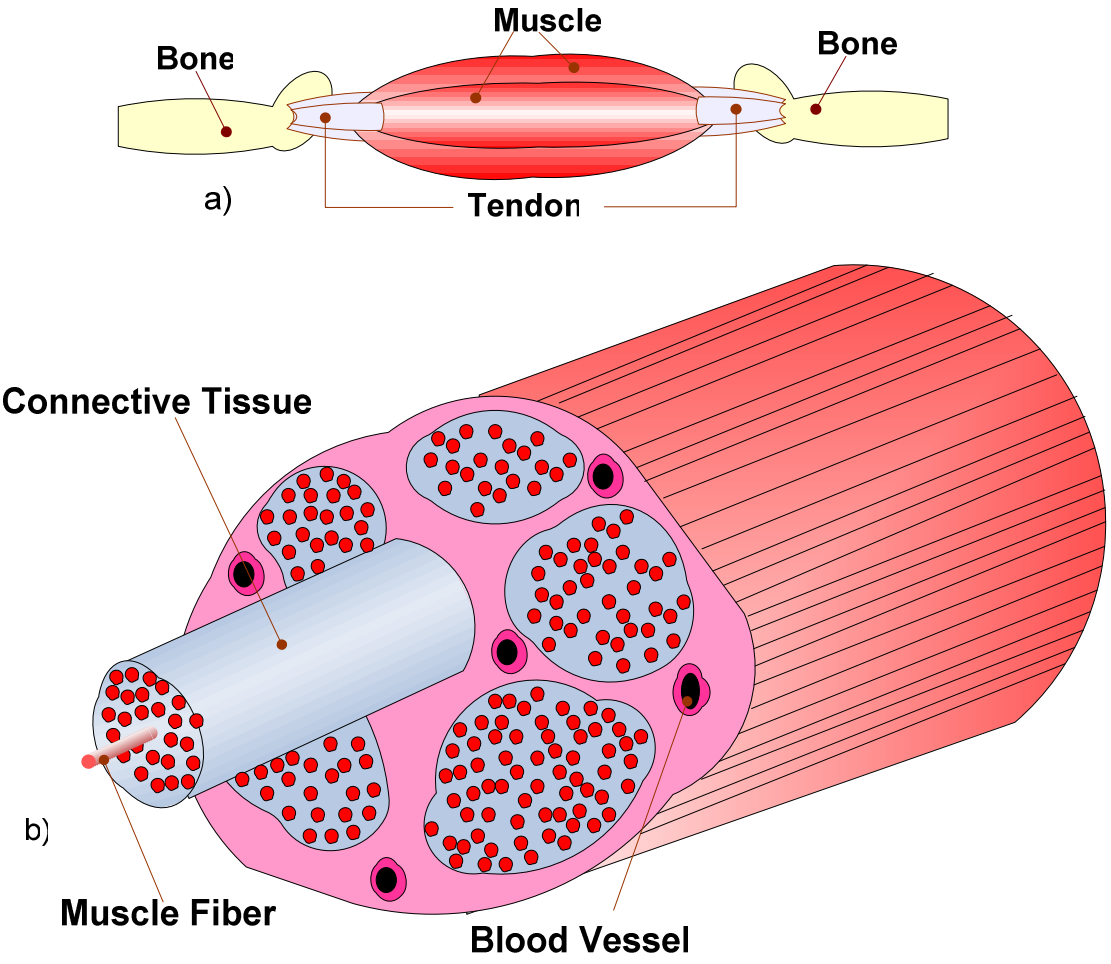

Figure 2.1 a) Representation of the attachment of skeletal muscle to bones via tendons. b) Organization of cylindrical muscle fibers

This is originated from the arrangement of numerous thick and thin filaments organized in the form of bundles of approximately 1 to 2 μm. These bundles are known as myofibrils. There are hundreds to thousands of myofibrils in a muscle fiber. These striations forming the repeating series of light and dark bands along the myofibrils are due to the arrangement of the thick and thin filaments in each myofibril. Since they contain contractile proteins such as myosin in the thick filaments and actin in the thin filaments, myofibrils are the contractile elements and consist of one unit of this repeating pattern called sarcomere bridges [7, 18].

Since they are anisotropic (i.e. they can polarize visible light) the dark bands are called A bands. This is the region where thick filaments are located. The A band is formed by the orderly parallel arrangement of these thick filaments. Since they are isotropic (non-polarizing), the light bands are called I bands bridges [7, 18, 21, 22].

Each sarcomere consists of two sets of thin filaments. One end of each thin filament is anchored to a network of interconnecting proteins known as Z-line. Z-line is a darker area located at the midline of I bands. The sarcomere is the region of a myofibril between two successive Z-lines bridges [7, 18].

I band is a light band that is located between the ends of the A bands of two adjacent sarcomeres. It contains the portions of the thin filaments that do not overlap thick filaments and is bisected by the Z-line bridges [7, 18].

There are additional stripes in the midsections of the A-band regions of each sarcomere which are called H-zone. There are relatively light bands. They correspond to the space between the thin filaments and only the central parts of the thick filaments are found in these regions bridges [7, 18].

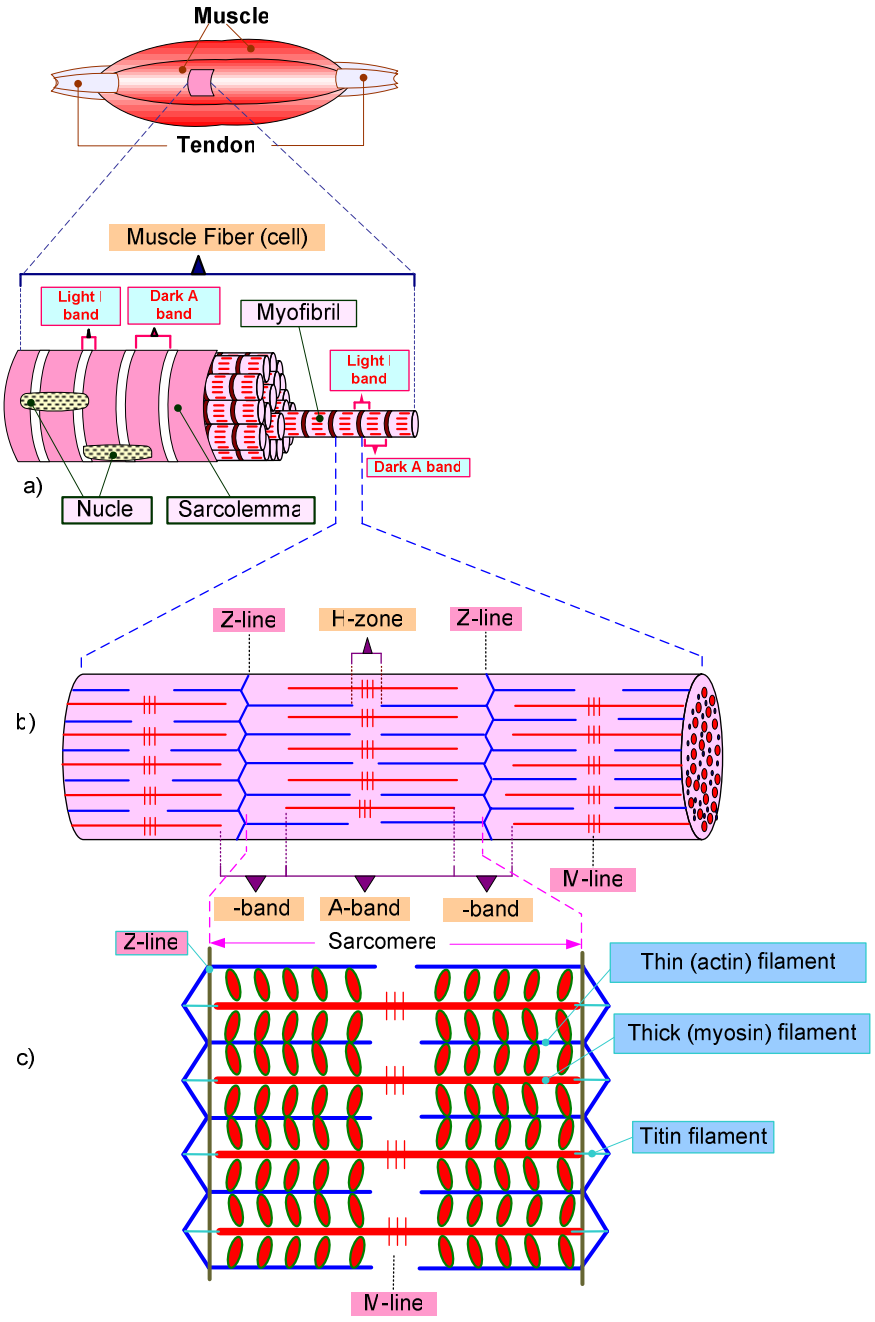

Figure 2.2 Microscopic anatomy of a skeletal muscle: a) A muscle fiber with its myofibrils; b) One myofibril with myofilaments responsible for the banding pattern c) One sarcomere containing thick and thin filaments.

The narrow dark band in the center of the H-zone is known as M-line corresponds to proteins linking together the central region of the thick filaments. The filaments composed of protein titin (connectin) which extend from the Z-line to M-line are linked to both M-line proteins and the thick filaments. This M-line linkage between the thick filaments and the titin filaments is responsible of maintaining the regular array of thick filaments centered in the middle of each sarcomere bridges [7, 18].

The thick filaments being 16 nm in diameter are primarily composed of the protein called myosin. It has a tail consisting heavy polypeptide chains and has two globular heads consisting light polypeptide chains as illustrated in Figure 2.3-a. Since they are responsible of linking thick and thin myofilaments during the contraction, the heads are also called cross-bridges [7, 18].

The myosin molecules form a bundle so that the tails constitute the central part of the thick filament and their heads protrude outward and in opposite directions at each end. A portion of a thick filament is shown in Figure 2.3-b [18].

The thin filaments are primarily composed of actin. The G-actin being the active sites binding the cross-bridges of the myosin filaments during the contraction are the polypeptide subunits of actin. The globular G-actin monomers are polymerized into fibrous actin or F-actin.

Two strands of F-actin forming a helical structure are the back-bone of thin filaments [18, 23]. There are regulatory proteins also in thin filaments. One of them is tropomyosin which is a rod-like protein. It forms a spiral beside F-actin. It plays role in the regulation of the interaction between actin and myosin during either contraction or relaxation. Other protein found in thin filaments is troponin. It is a three-polypeptide complex.

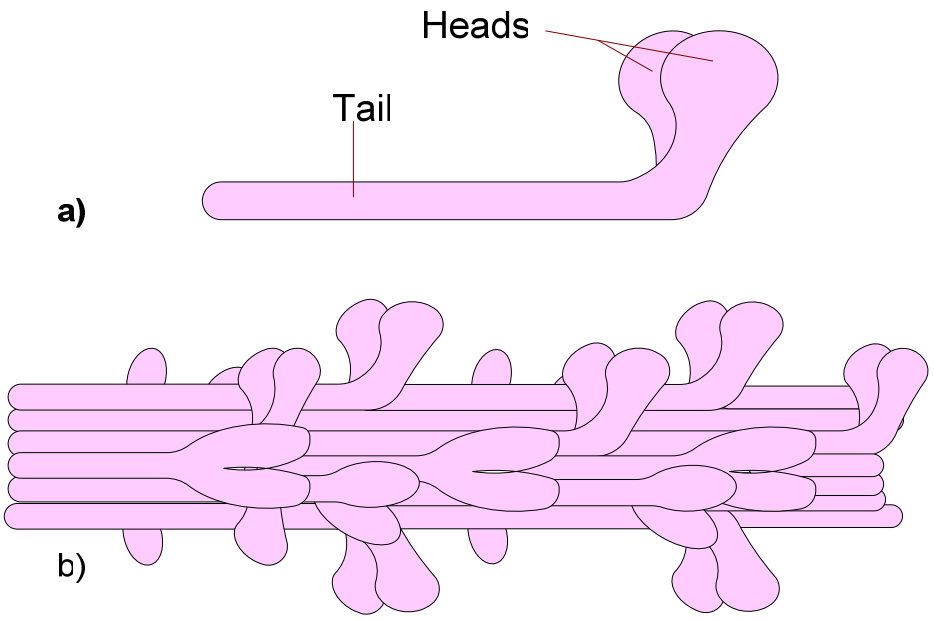

Figure 2.3 a) Myosin Molecule b) Portion of a thick filament

These polypeptides complex are TnI which is an inhibitory subunit binding to actin, TnT binding to tropomyosin helps troponin to position on actin and TnC that binds calcium ions. Like tropomyosin, troponin helps also to control the myosin-actin interaction involved in contraction [18]. The structure of thin filaments is represented in Figure 2.4.

The endoplasmic reticulum of a muscle fiber is referred as Sarcoplasmic Reticulum (SR). The myofibril is surrounded by the interconnecting tubules of the sarcoplasmic reticulum. Most of these tubules lie longitudinally along the myofibril. On the other hand, there are larger channels at the end of each segment known as terminal cysternae. These channels store calcium which are released when the membrane is excited [7, 18].

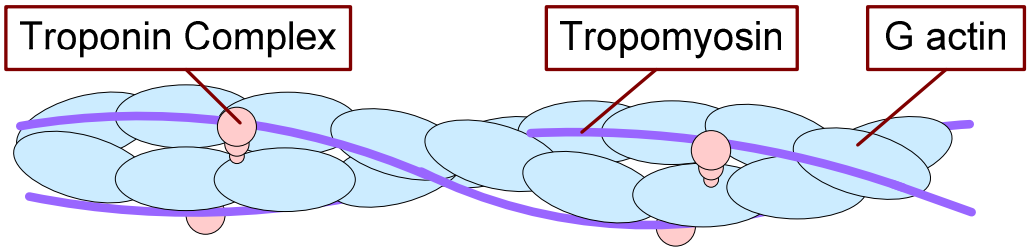

Figure 2.4 Portion of a thin filament

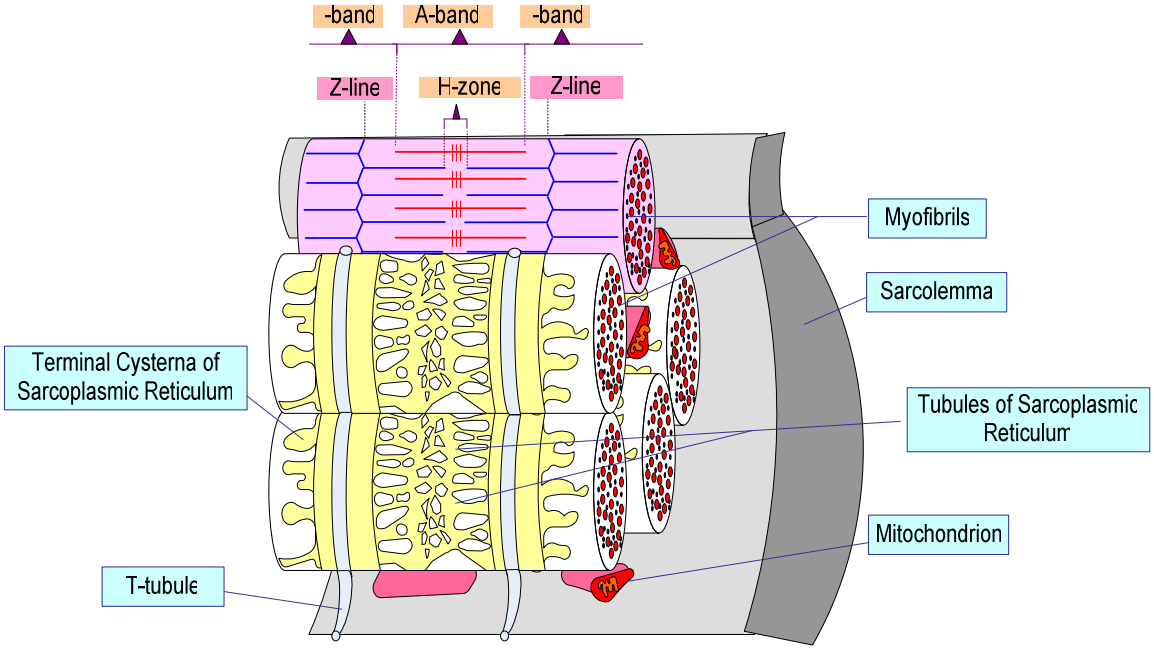

Figure 2.5 Relationship between the sarcoplasmic reticulum and T-tubules and the myofibrils

The sarcolemma penetrates into the muscle fiber forming a hollow tube at the boundary between A-band and I-band. These tubes are called transverse tubules or T-tubules. They have continuous lumen with extracellular space. They help to the propagation of the muscle action potential to the deepest part of the muscle cell [7, 18].

The relationship between the sarcoplasmic reticulum and T-tubules and the myofibrils are illustrated in Figure 2.5.

2.3 Physiological Processes of the Skeletal Muscle Contraction:

2.3.1 Basic Motor Control Mechanism

The neurons involved in controlling skeletal muscles are organized in a functional hierarchical fashion. Each level of the hierarchy has a certain task in motor control. This hierarchical organization is shown in Figure 2.6. To initiate an action, a decision should be made in the highest level of this organization. Although the knowledge about this level is limited, it is assumed that it includes many regions of the brain comprising those involved in memory, emotions and motivation [7].

Information from this level is transmitted to parts of the brain constituting the middle level of the motor control hierarchy. This level is responsible of determining the movement needed to establish the decided action and of maintaining the posture.

The anatomic structures constituting the middle level of the motor control hierarchy are located at the sensorimotor cortex, cerebellum, subcortical nuclei and brainstem. They integrate afferent information coming from receptors in the muscles, tendons, joints, skin, vestibular apparatus and eyes with the signals coming from the highest level of the motor control hierarchy to perform the decided movements known as motor program.

The information about the motor program is conveyed through the descending pathways to the local level which is the lowest level of the motor control hierarchy. The descendant pathways are classified as brainstem pathways and corticospinal pathways. The brainstem pathways are also called extrapyramidal system. Axons from neurons in the brainstem form pathways descending into spinal cord in order to influence motor neurons. They play role in the control of upright posture, balance and walking [7, 18]. The corticopinal pathways are also known as pyramidal system. The nerve fibers of the corticospinal pathways orginate from the sensorimotor cortex and terminate in the spinal cord. The corticospinal fibers are the source of control for voluntary movement of the skeletal muscles [7, 18].

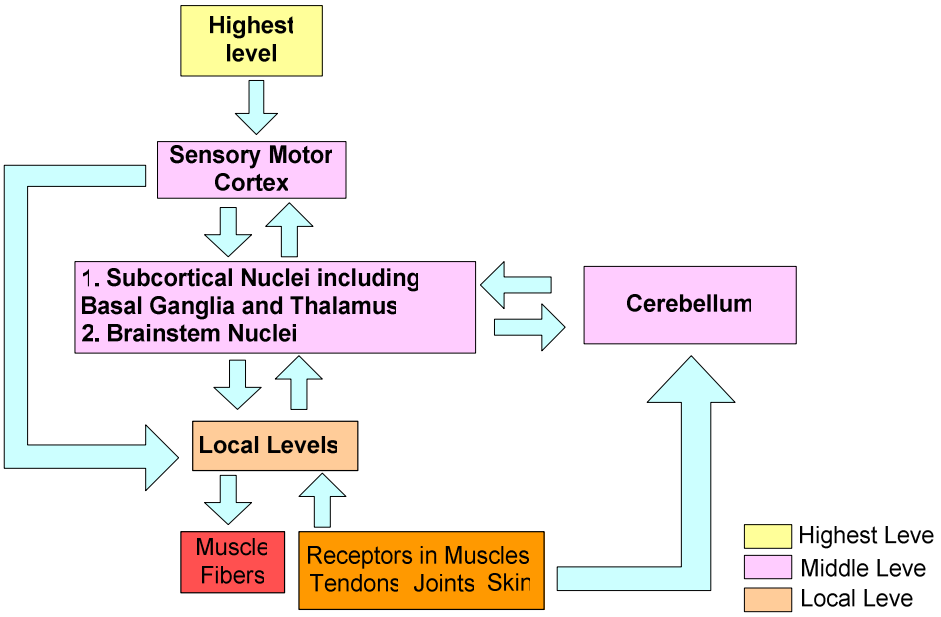

Figure 2.6. Functional hierarchical organization of the neural system controlling the body movement

In the local level which is the lowest level of the motor control hierarchy, the corticospinal fibers have synapses with the anterior horn cells which are the primary motor neurons [7, 18, 24]. The information about the motor program is conveyed

through these pathways in the form of action potentials [25, 26]. These pathways are illustrated in Figure 2.7.

The last component of the motor control hierarchy is the motor neuron that transmits the electrical pulse in the form of action potential initiating the muscle contraction. The primary motor neurons are the anterior horn cells and they are located in the ventral gray matter of the spinal cord. Their projections first pass through the white matter of the spinal cord before exiting as the motor roots. Their axons become the motor fiber of the peripheral nerves with myelin sheath that innervate skeletal muscles [24, 26].

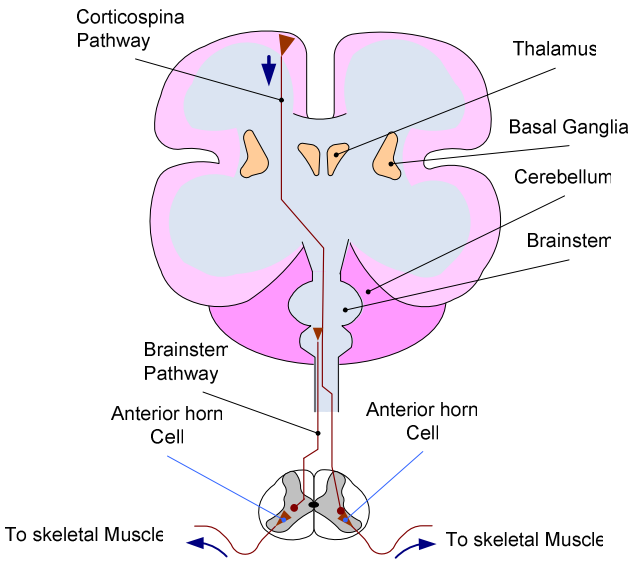

Figure 2.7 The descending pathways that convey information about the motor program (Arrows indicate direction of action potential propagation).

2.3.2 Neuromuscular Junction (NMJ)

The information about the motor program is transmitted from the local level of the motor control hierarchy to the muscle fiber via the axon of the motor neuron in order to initiate the contraction of skeletal muscle [19]. Each muscle fiber is innervated by the axon of only one motor neuron [20]. The axon of each motor neuron divides into many branches when it reaches to the surface a muscle and become unmyelinated [7, 18]. The connection between an axon terminal and a muscle fiber is represented in Figure 2.8-a.

In the region of contact, the muscle fiber is modified to form the motor end-plate that contains mitochondria and nuclei [27]. The junction of axon terminal of a motor neuron with the motor end plate called neuromuscular junction [7, 18]. There is a space about 500 Å wide, between the nerve terminal and the muscle cells, which is called synaptic cleft [4, 27].

The neuromuscular junction is a chemical synapse ensuring the functional connection between the motor neuron and the target skeletal muscle [19]. At these junctions, the axons terminals contain large numbers of synaptic vesicles about 500 Å in diameter concentrated in regions opposite the folds in the postsynaptic membrane. These vesicles contain a neurotransmitter called acetylcholine (ACh) [27]. The neuromuscular junction is illustrated in Figure 2.8-b.

When an action potential transmitted by the motor neuron arrives at an axon terminal, it depolarizes the nerve plasma and then opens voltage-sensitive calcium channels allowing calcium ions to diffuse into the axon terminal from the extracellular fluid. Calcium binds to proteins enabling the membrane surrounding the acetylcholine-containing vesicles and helps them to fuse with nerve plasma membrane in order to

release acetylcholine into the synaptic cleft between axon terminal and the motor end plate [7, 18].

The motor end plate contains numerous smaller folds that provide a large surface area for many nicotinic ACh receptors being located there [4, 18]. The ACh molecules that diffuse through the synaptic cleft, then bind to nicotinic acetylcholine receptors on the sarcolemma. Hence, the ion gates opens leading to permeability changes in the sarcolemma. This results in the depolarization of the sarcolemma [4, 18]. Initially, an end-plate potential (EPP) occurs due to the localized depolarization of the sarcolemma at the motor end plate [27]. Provided that sufficient neurotransmitter is released from the axonal terminal of the motor neuron, EPP will exceed a threshold to activate adjacent voltage-gated ion channels in the adjacent sarcolemma yielding an action potential propagating along the sarcolemma [5, 27, 28]. The release of acetylcholine is represented in Figure 2.9.

Since the membrane depolarization produced in the muscle by the nerve action potential will be reduced by perhaps 1/100 times the depolarization the nerve induces itself, direct transfer from of electrical activity from nerve to muscle cannot take place. Therefore, the mechanism whereby the motor neuron stimulates its target muscle depends on a chemically mediated junction rather being electrical. The role of neuromuscular junction is being an impedance transformer from high impedance nerve to low impedance muscle [5, 28].

After a few milliseconds, the acetylcholine is broken down by an enzyme called acetylcholinesterase (AChE) in order to prevent the continued muscle fiber contraction in the absence of additional stimulation coming from the motor neuron [4, 18].

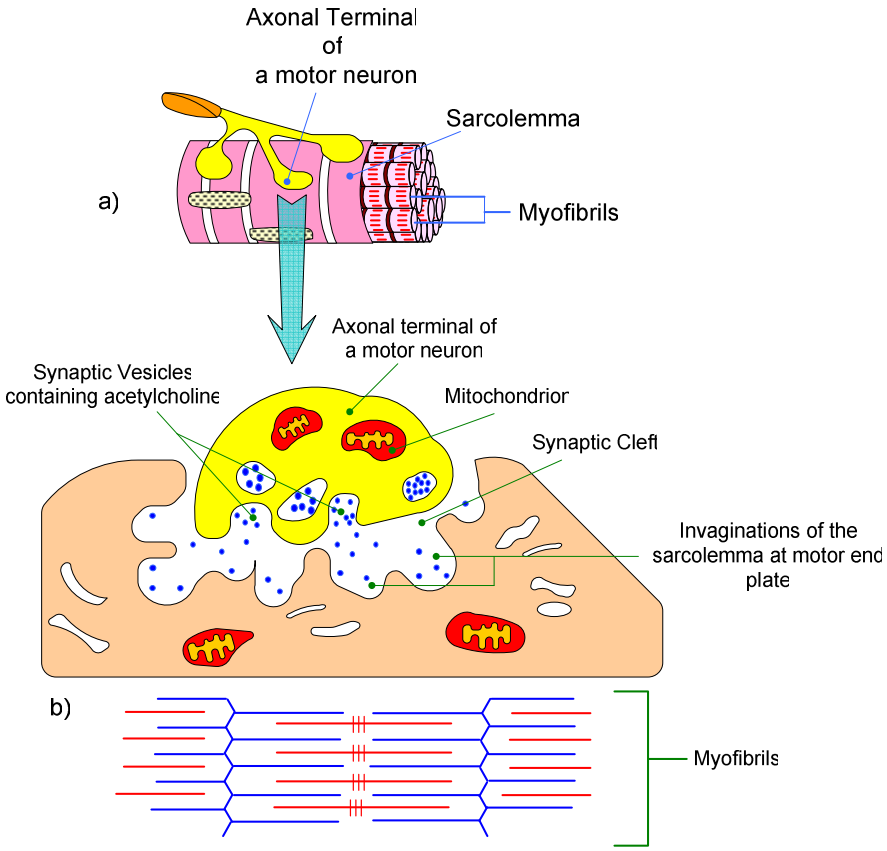

Figure 2.8. a) Axonal ending of a motor neuron forming a neuromuscular junction with a muscle fiber. b) Neuromuscular junction.

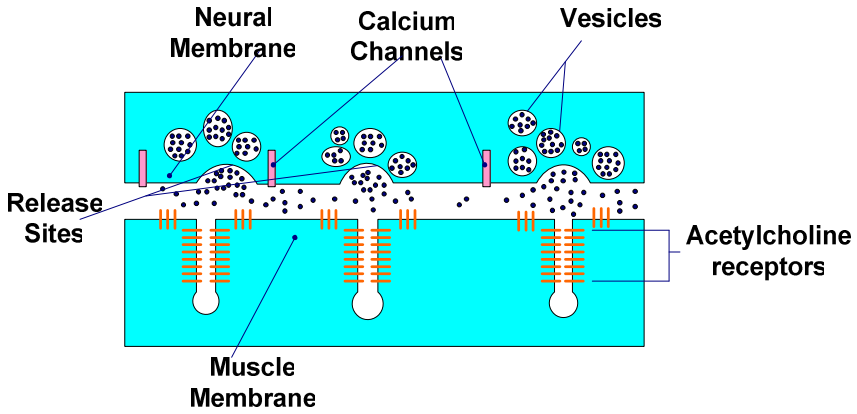

Figure 2.9. Schematic representation of the release of acetylcholine.

2.3.3 Excitation-Contraction Coupling

The excitation-contraction coupling comprises the events related with the conversion of the chemical energy provided by the adenosine tri phosphate (ATP) molecules into mechanical force produced by the filaments of the myofibrils.

Since the sarcoplasma is an excitable membrane an action potential is generated in the post-synaptic part of the neuromuscular junction following the chemical transmission of the signals coming from the motor neuron through this neuromuscular junction [4, 7] [18].

This action potential is propagated not only through the skeletal muscle membrane referred as sarcoplasma but also along the infoldings of the surface membrane called tranverse or T-tubules [7]. This electrical activity does not directly act on the contractile proteins. However, it contributes to the initiation of the contractile mechanism by increasing cytosolic calcium concentration. This is established by the activation of the voltage-gated calcium channels located in membrane of the T tubules [7, 19, 20].

These channels are in close apposition to the calcium release proteins in the Sarcoplasmic Reticulum and their conformational changes due to the depolarization of T-tubule trigger the release of calcium stored in the terminal cisternae of the Sarcoplasmic Reticulum into the muscle cytoplasm [20].

In a muscle fiber with resting state, the concentration of calcium ions in the cytoplasm surrounding thick and thin filaments is about 10^{-7} molar which is not sufficient to elicit contraction [4, 7]. In this state, since very few of the calcium-binding sites are occupied,

the cross-bridge interaction between thin and thick filaments resulting in contraction is blocked by tropomyosin maintaining the muscle in a resting state [4, 7].

Following the depolarization of the T-tubules by the action potential generated at the post-synaptic part of neuromuscular junction and propagated along the sarcolemma, an increase in the concentration of calcium ions in the cytoplasm surrounding the myofibrils occurs reaching approximately 2×10^{-4} molar [4, 7].

The elevation of the intracellular calcium activates contractile proteins [20]. These calcium ions diffuse passively among the myofilaments and reversibly bind to the troponin C causing a conformational change in tropomyosin molecule. This results in removing the blocking effect of the tropomyosin and thus allowing the cross-bridge interaction [4, 7, 23].

The contraction will continue until calcium is removed from troponin [23]. Thus, the calcium concentration in the cytoplasm should be lowered back to pre-release level. The membrane of the sarcoplasmic reticulum possesses active-transport proteins referred as Ca-ATPases that pump calcium ions from the cytoplasm back into the sarcoplasmic reticulum [7]. When the re-uptake of calcium ions is achieved relaxation occurs [20]. The stages of the excitation-contraction coupling are demonstrated in Figure 2.10 and it is schematized in Figure 2.11.

2.3.4 Contractile Process

The contraction is achieved through the interaction between the thick and thin filaments of the muscle fiber. In the presence of calcium, cross-bridge interaction

between actin and myosin filaments take place to establish muscle contraction and to convert the chemical energy supplied by ATP molecules into the mechanical work [4, 5, 27].

2.3.4.1 Sliding Filament Mechanism:

The muscle contraction occurs by a sliding filament mechanism [4]. In this mechanism, actin filaments slide inward among the myosin filaments. This is achieved by mechanical forces generated by interaction of the cross-bridges from the myosin filaments with actin filaments [4, 27].

Hence, a shortening of a skeletal muscle fiber is produced [7]. During this shortening the thin filaments slide centrally, the Z-discs to which the thin filaments are attached are pulled toward the thick filaments resulting in the reduction of the distance between successive Z lines. I bands shorten, the H zones disappear, and the A bands move closer together [18]. Even though the sarcomeres shorten (i.e. the distance between the successive Z lines), there is no change in the length of either the thick or thin filaments [7]. Sliding Filament Mechanism is illustrated in Figure 2.12 and in Figure 2.13.

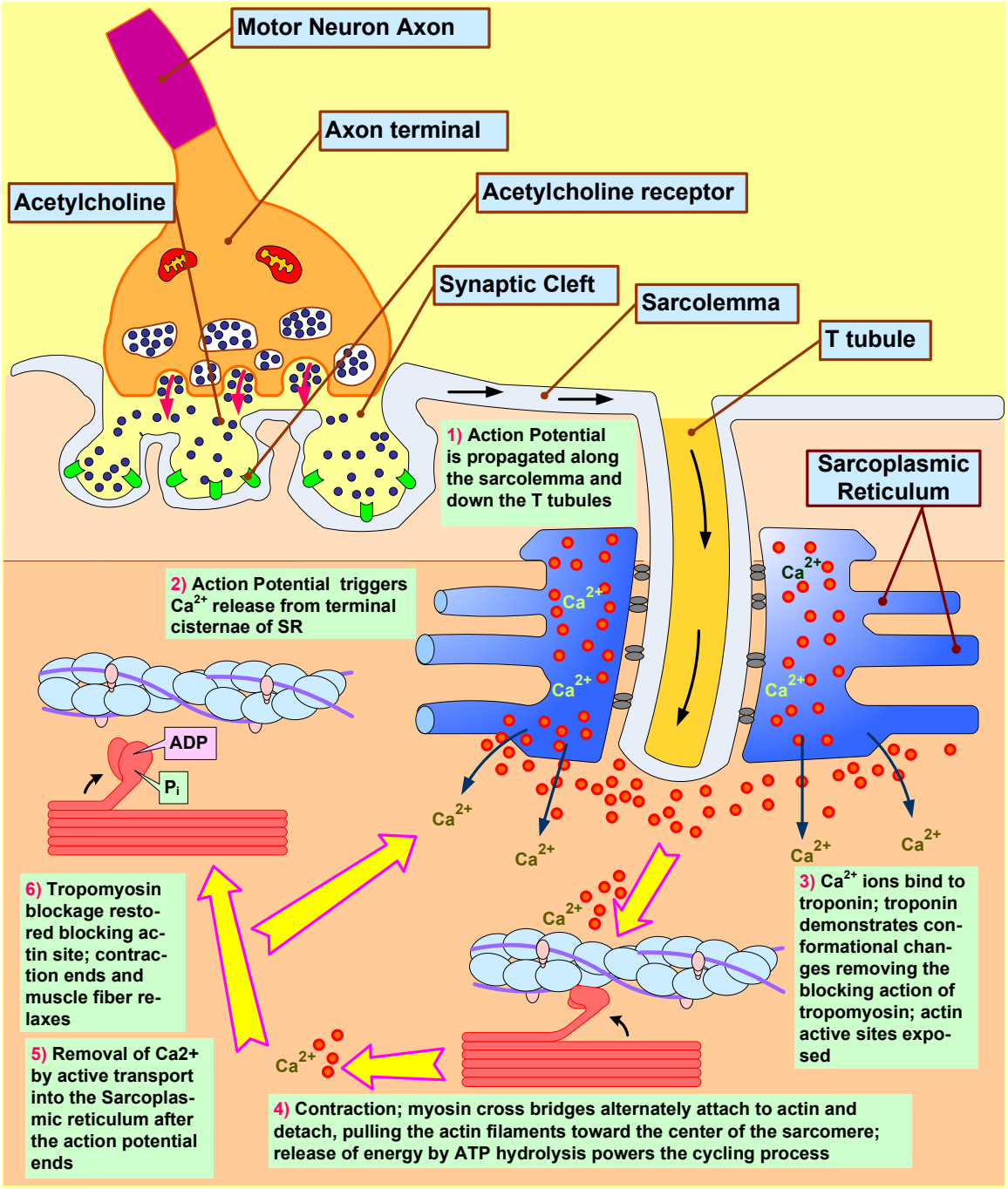

Figure 2.10 Stage of the Excitation-Contraction Coupling

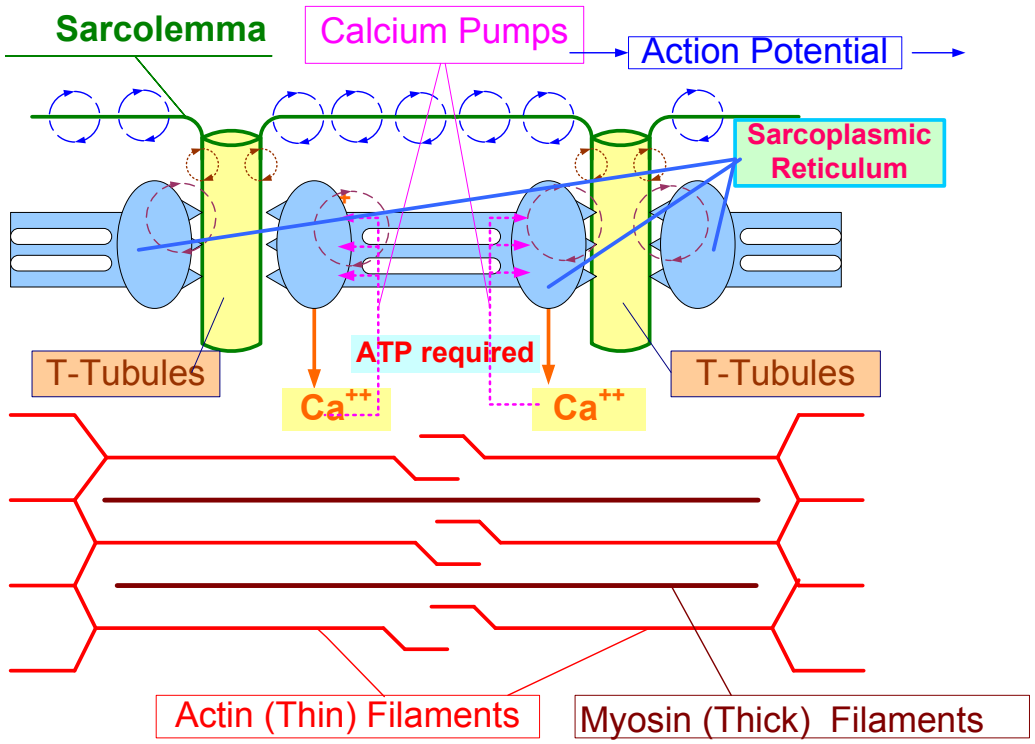

Figure 2.11 Schematic illustration of the Excitation-Contraction Coupling

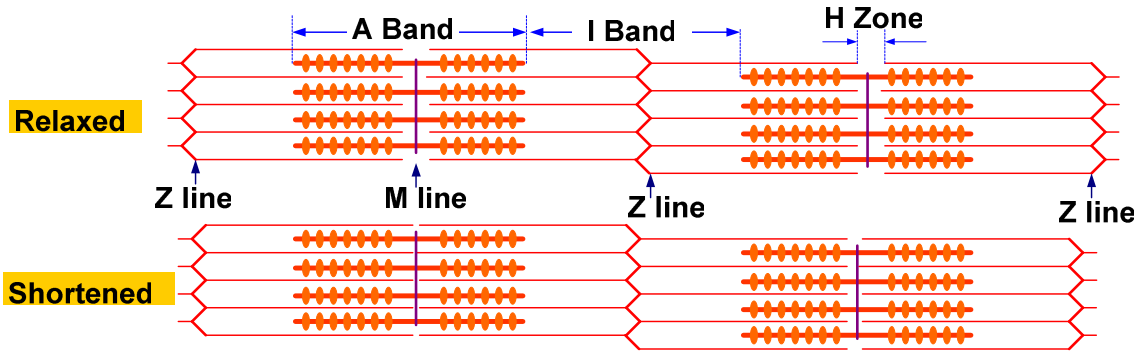

Figure 2.12. The sliding of thick filaments past overlapping thin filaments.

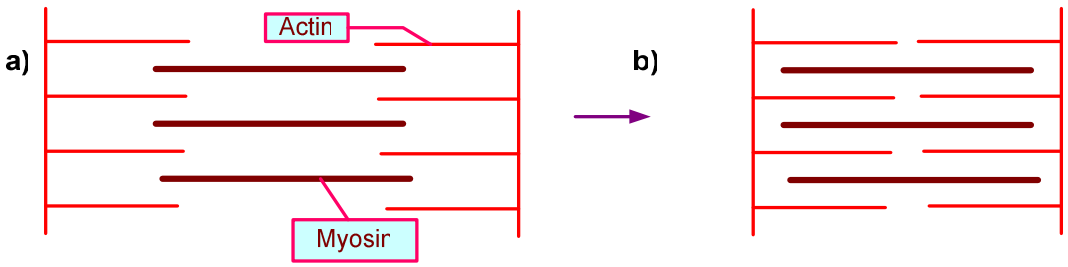

Figure 2.13 Schematic represention of the Sliding Filament Mechanism.

2.3.4.2 Interaction of Myosin and Actin Filaments:

Before the contraction initiates, the active sites of the F-actin filament are physically covered by troponin-tropomyosin complex to inhibit the interaction between the head of the myosin molecules and the active sites of the actin molecules as the intracellular calcium concentration levels are low [4]. The head myosin molecules have two sites which one of them is the active-binding site interacting with the F-actin molecule and the other one is the Myosin-ATPase site [7, 22]. These are shown in Figure 2.14.

At low intracellular calcium levels, the muscle is relaxed and the active myosin binding sites are physically covered by tropomyosin molecules to inhibit the actin-myosin interaction [4, 18]. After a nerve impulse coming from motor neuron, an increase in calcium ions caused by the calcium release from the terminal cisternae of the Sarcoplasmic Reticulum within the muscle cell occurs to activate the actin-myosin interaction [18].

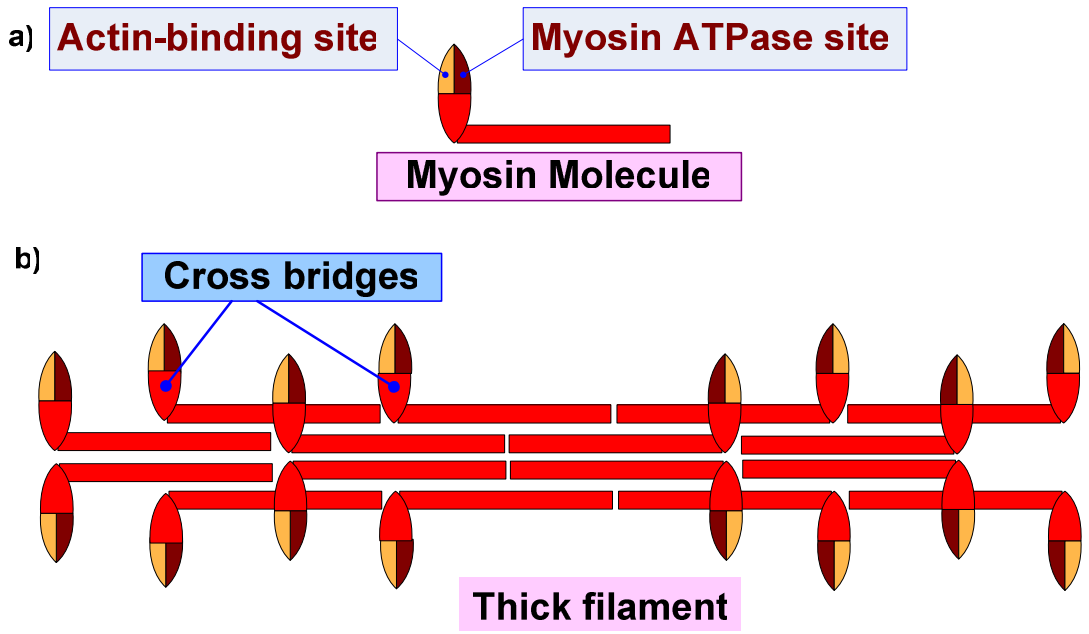

Figure 2.14 a) The schematic structure of the myosin molecule. **b)** The globular heads of myosin constitute cross bridges and the myosin tails are located in the core of the myosin (thick) filament.

The sequence of events occurring during this interaction to establish the contraction can be summarized as follows [4, 7, 18];

i. Before the contraction begins, the head of myosin binds with ATP. Since work is performed for the contraction, energy is required. It is provided by splitting ATP molecule into ADP and Pi through the hydrolysis of ATP by the ATPase activity of the myosin head. These hydrolysis products are left bound to the head. The head is extended perpendicularly or cocked toward the actin however it is not yet attached.
ii. When the troponin-tropomyosin complex binds with calcium ions released from the Sacrcoplasmic Reticulum, the blockade of this complex is removed and the active sites of the actin filaments are exposed. Then, the actin-binding sites of the myosin heads can bind these active sites. This process is shown in Figure 2.15.
iii. Conformational change in the myosin head leading the head to tilt toward the arm occurs due to the bond between the head of the cross-bridge and the active site of the

actin filament. This results in power stroke to pull the actin filament toward the center of the sarcomere as shown in Figure 2.16. As a result, swiveling motion of the cross-bridges is established for the displacement of the filaments explained by the walk-along (or ratched theory) illustrated in Figure 2.17. The energy required for the power stroke is ensured form the cleavage of ATP and is already stored in the cross-bridges as if it is a cocked spring.

iv. The ADP and Pi molecules attached to the myosin head are released after it tilts toward the cross-bridge. A new ATP molecule binds to the head of myosin and the head is detached form the active site of the actin.
v. After the head is detached from the actin molecule, ATP is cleaved again to begin next cycle of the contraction.
vi. The cocked head bind with this stored energy supplied by the cleavage of ATP to the new active site of the actin filament to generate a new power stroke.

The contraction ceases when the intracellular calcium is pumped back to the Sarcoplasmic Reticulum through the active transport by means of the Ca-ATP'ases in order to remove calcium from the troponin [7].

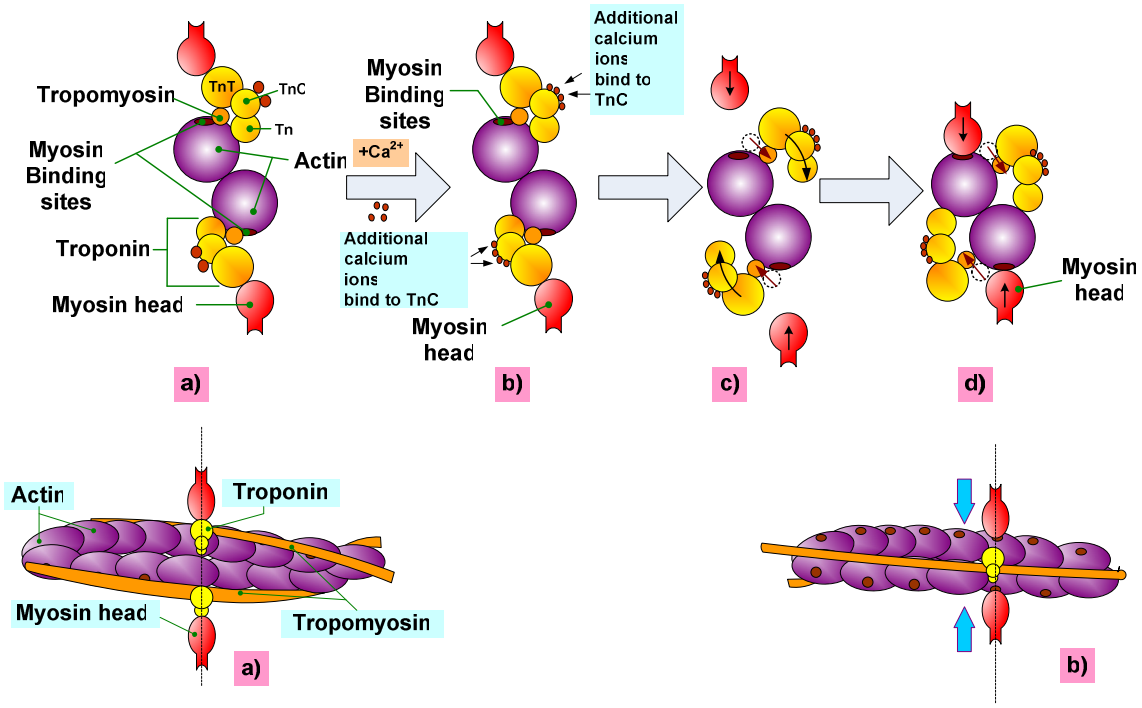

Figure 2.15 The actin-myosin interaction in the presence of calcium ions; **a)** Blockade of tropomyosin at low intracellular Ca^{2+} concentrations **b)** Binding additional calcium to troponin C (TnC) at higher intracellular Ca^{2+} concentrations **c)** Conformational changes in troponin moving the tropomyosin away from the active sites of actin **d)** Myosin heads bind with the active sites of actin initiating the sliding mechanism of the contraction.

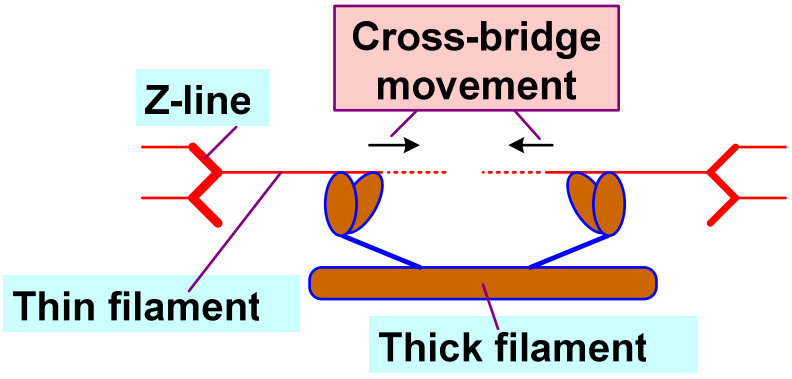

Figure 2.16 Conformational change in cross-bridges of the thick (myosin) filaments to pull the thin (actin) filament toward the center of the sarcomere

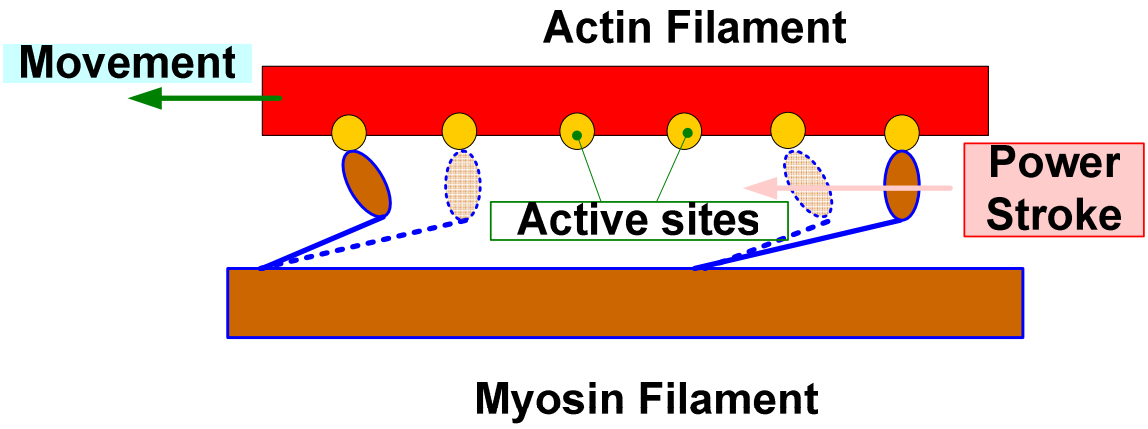

Figure 2.17 Walk-along mechanism for the contraction of the muscle.

2.4 Electrophysiological Characteristics of the Skeletal Muscle:

Skeletal muscle is one of the excitable tissues like the nerves. They can generate electrical signals called action potential when they are stimulated internally or externally. This is the origin of the electrophysiological aspects of the skeletal muscle. These electrophysiological aspects form the basis of the electrodiagnostic studies such as Electromyography (EMG). EMG is based on the detection of Motor Unit Action Potential (MUAP) generated by the Motor Units in various sizes in a certain muscle.

2.4.1 Motor Unit

A typical muscle is controlled by about a hundred large motor neurons whose cell bodies lie in a distinct cluster called a motor nucleus in the spinal cord or in brain stem. The axon of each motor neuron exits the spinal cord through a ventral root or through a

cranial nerve from the brain stem. It proceeds in the form of smaller branches of peripheral nerves until it enters the muscle it controls. Then, it branches widely to innervate from 100 to 1000 muscle fibers scattered over a substantial part of the muscle. The number of muscle fibers innervated by the same motor neuron is termed innervation ratio and can considerably vary for different muscles. It ranges from 3:1 in extrinsic eye muscles which require fine gradation of movement to 30:1 to more than 2000:1 in limb muscles establishing only coarse movement [1, 6, 26]. The muscle fibers innervated by a single motor neuron and the motor neuron itself are referred as a motor unit. Motor Unit (MU) is the basic structural and functional unit of skeletal muscles [8]. It is the last common path playing role in motor functions. Each muscle fiber is innervated by only one motor unit. [19].

The action potential conveyed through the motor neuron always results in depolarization of all muscle fibers of the motor unit synchronously creating a MUAP [19].

2.4.1.1 Structure of the Motor Unit:

A motor neuron system consists of anterior horn cells, its axons consisting radix, plexus and peripheral nerves and neuromuscular synapses. Anterior horn cells are the primary motor neurons. They are located in the ventral gray matter of the spinal cord. The axons of these cells become the motor fibers in peripheral nerves. These axons are effectively insulated with myelin. Depolarization occurs by way of saltatory conduction and only at each node of Ranvier. Since much less nerve membrane has to be depolarized, less time is required. Therefore, the conduction velocity is increased. The salutatory conduction is represented in Figure 2.18. The myelinated nerve fibers conduct in the range from 35 to 75 m/s. The Motor Unit is defined as one axon, its anterior horn cell, and all connected muscle fibers and neuromuscular junctions [5, 24, 27, 29, 30].

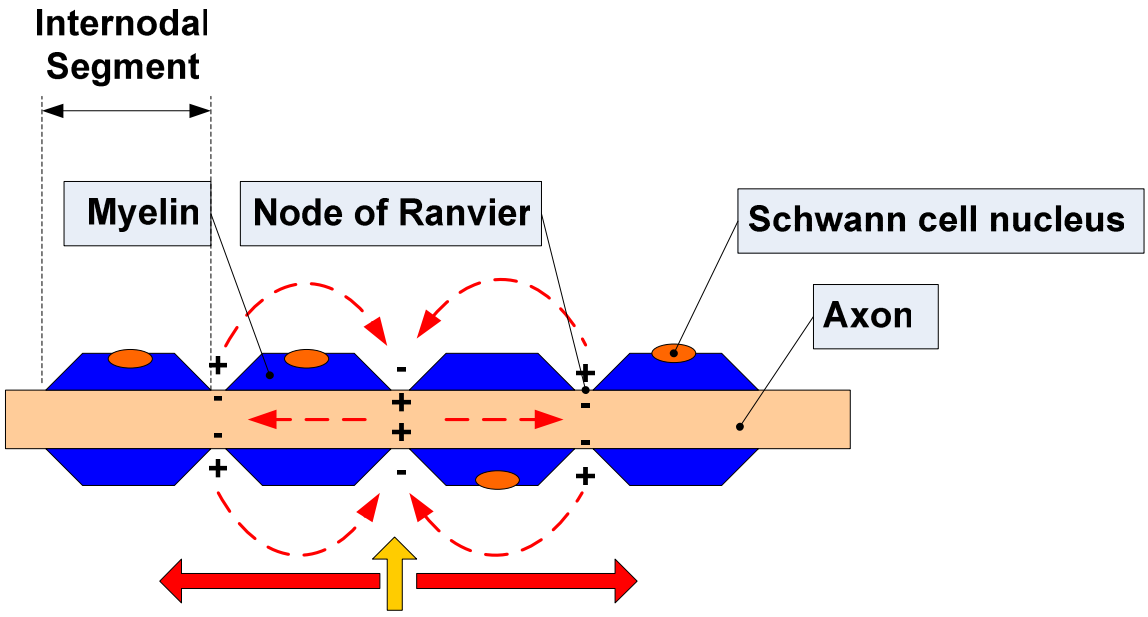

Figure 2.18 Saltatory conduction along the myelinated fiber. The myelin sheath insulates the internodal segment. Depolarization occurs at the node of Ranvier. The current flows between intracellular and extracellular fluid. A local current is generated at one node depolarizes the axis cylinder at the adjacent nodes on either side to transmit the impulse in both directions.

The motor unit and its components such as one axon, its anterior horn cell, all connected muscle fibers and neuromuscular junctions are represented in Figure 2.19. The cross section of muscle occupied by a motor unit is called the motor unit territory (MUT) [3].

The muscle fibers belonging to a single motor unit are intermingled (i.e. scattered throughout the muscle) in a mosaic pattern, rather lying adjacent to each other [7, 19, 20, 31].

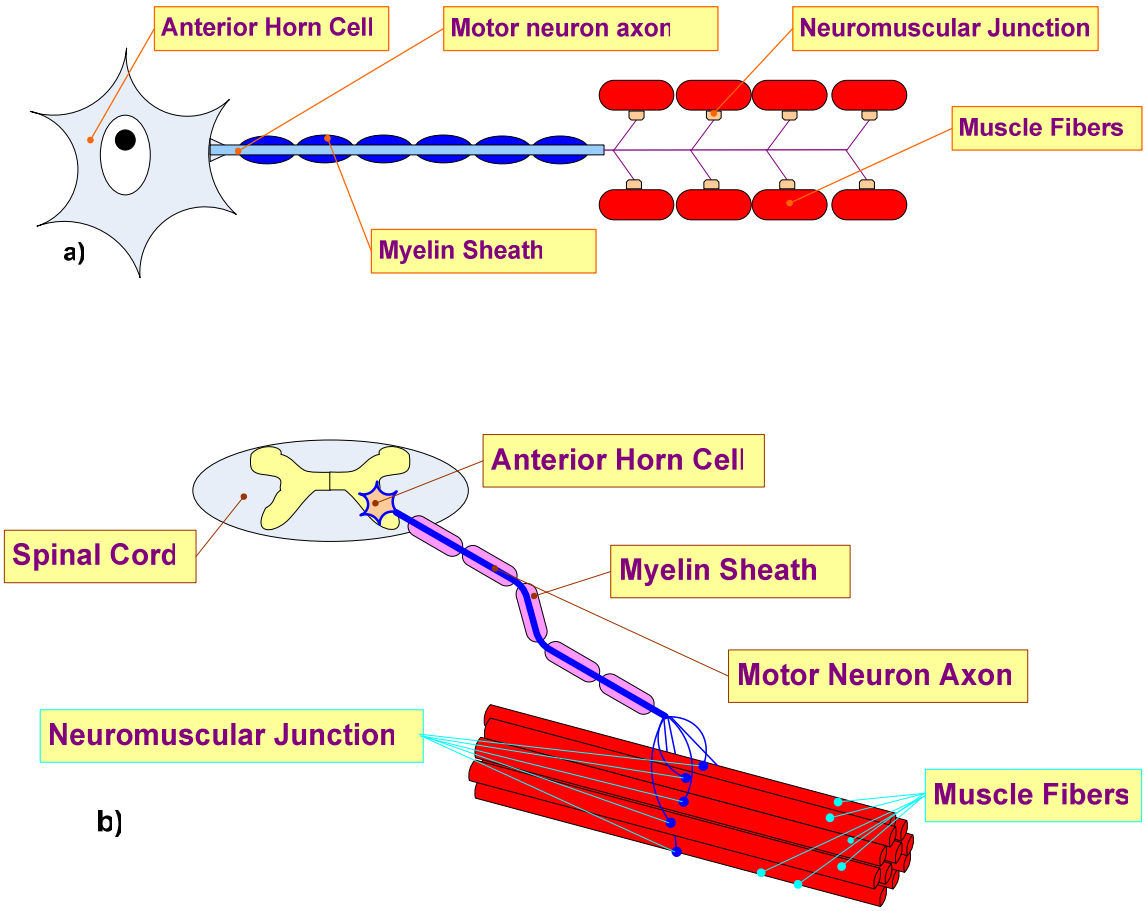

Figure 2.19 The motor unit: **a)** Simplified schematic representation of a motor unit **b)** Representation of the components of a motor unit (anterior horn cell, motor neuron with its axon and its myelin sheaths, neuromuscular junction and the muscle fibers)

2.4.1.2 Types of Motor Units:

The muscle fibers innervated by one motor neuron in a motor unit consist of the identical fibers [26]. Thus, the properties of the muscle fibers of a motor unit have a great importance in the determination of the motor unit types. On the other hand, some properties of the components should be considered in classifying the motor unit types.

The types of the motor units are classified according to the histochemical and metabolic properties of the muscles fibers, to mechanical and contractile properties of the muscle fibers, to electrophysiological properties of the muscle fibers, to electrical properties of the motor neuron, to electrophysiological properties of the axons and of the neuromuscular junction of the motor unit [2, 6, 19]. This classification is achieved most commonly upon the histochemical and metabolic properties of the muscle fibers such and/or upon the contractile and mechanical properties (e.g. twitch properties and the fatigability) of the muscle fibers belonging to a motor unit [2, 19, 20].

The muscle fibers are divided into three types according to their histochemical staining and to their metabolic properties such as type I, type IIa and type IIb [22]. After preincubation at pH 4.6, type I fibers stain dark, type IIa fibers remain unstained and type IIb fibers are moderately stained [2]. The histochemical properties of muscle fibers are summarized in Table 2.1.

The muscle fibers can be classified according to their twitch contraction properties and fatigability such as slow twitch (S-type), fast-twitch fatigue resistant (FR-type) and fast-twitch fatigable (FF) [2, 20]. The twitch contraction property can be defined as the speed of the response to motor neuron action potential as the peak force or the peak tension generated by the muscle fiber and the relaxation time [20]. It is shown in Figure 2.20.

Since the contractile force of a motor unit depends on the force-generating capabilities of its fiber type multiplied by the numbers of its fibers innervated by the motor neuron the motor units are also classified as type I (S-type), type IIa (FR-type) and type IIb (FF-type) [19].

Table 2.1.

Histochemical properties of Muscle Fibers

	Type I	Type IIa	Type IIb
ATPase (pH 4.3)	Strong	Weak	Weak
Succinic dehydrogenase	Strong	Strong	Weak
NADH reductase	Strong	Intermediate	Weak
Glycogen stores	Low	High	High
Myophosphorylase	Low	High	High
Myoglobulin	High	Low	Low
Lipid globules	Many	Many	Few

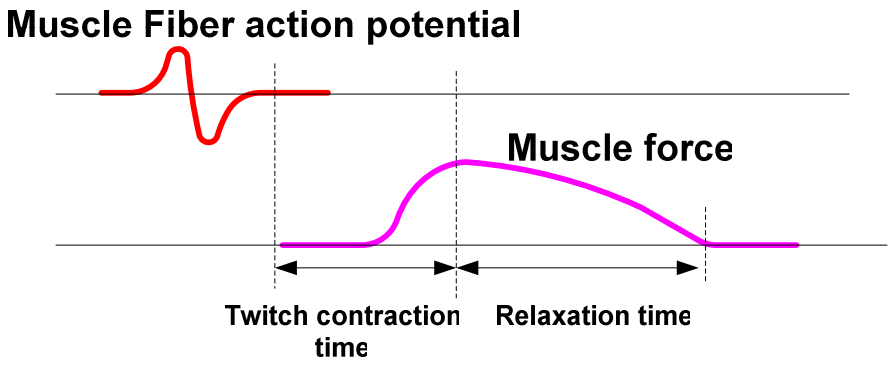

Figure 2.20 The twitch contraction properties are determined by the latency to reach peak force and the relaxation time.

Type I (S-type) motor units possess type I (Slow-twitch) muscle fibers. The force produced by these fibers rises and falls relatively slow in response to an action potential generated in the sarcolemma of the muscle fiber [7]. They have numerous mitochondria with a high capacity for oxidative phosphorylation and oxidative enzymes for

synthesizing ATP. So, these fibers are also classified as oxidative fibers. Since the synthesis of ATP depends on the blood flow to deliver oxygen and fuel molecules such as glucose and fatty acids, these fibers are surrounded by blood vessels. Since they consist of large amounts of myoglobulin being an oxygen-binding heme protein which increases the rate of oxygen diffusion and providing storage of oxygen from the circulation, they have dark-red color and are referred as red muscle fiber [7, 20]. The oxidative (aerobic) metabolic property makes these fibers as the most fatigue-resistant fibers [19, 20]. On the other hand, they contain myosin with lower ATP'ase activity [7]. They produce less contractile force because they are the smallest fibers in diameter and possess less contractile elements. The motor neurons consisted in type I (S-type) motor units have also smaller axons innervating fewer, thinner fibers with the metabolic properties mentioned above and lower nerve conduction velocity [2, 6, 19]. These motor units are activated at small forces and are recruited at lower force levels [2, 6].

Type IIa (FR-type) motor units consist of type IIa muscle fibers. They have faster twitch and faster relaxation in response to the action potential generated in sarcolemma compared to type I fibers. They possess moderate number of mitochondria and are able to use both oxidative metabolism and glycolysis to ensure energy for the contraction [19]. Therefore, they are relatively resistant to fatigue [7, 19]. On the other hand, they have relatively high ATP'ase activity. Since they contain lower myoglobin than type I fibers, they are pale when compared to type I fiber, however, they are darker than type IIb fibers. Since they have larger fibers and more contractile elements, they produce relatively more contractile force compared to type-I. The motor neurons of type IIa motor units have intermediate axons in size with moderate nerve conduction velocity. They are activated by intermediate forces and hence are recruited at moderate force levels. [2, 6, 29].

Type IIb (FF-type) motor units consist of type IIb muscle fibers. They have the fastest twitch and fastest relaxation as a response to muscle fiber action potential. They have sparse mitochondria and a high concentration of glycolytic enzymes and large store

of glycogen [7, 19]. The energy required for the contraction is provided by the glycolysis of the glycogen to phosphorylate ADP rapidly and by converting the glycogen into lactic acid [2, 6, 7]. This rapid depletion of glycogen and accumulation of lactic acid limit these fibers to brief burst of force [19]. Hence they are highly fatigable [7, 19, 20]. Due to their glycolytic (anaerobic) metabolic properties, they are surrounded by few blood vessels and contain little myoglobin and they are pale colored [7, 20]. Since type IIb muscle fibers are large fibers and they produce high specific forces [19, 20]. They have highest myosin-ATP'ase activity [7]. Type IIb motor units have large cell bodies and large-diameter axons conducting motor neuron action potentials at higher velocities [19].

The metabolic, mechanical and electrical properties of three types of motor units are summarized in Table 2.2.

Table 2.2.

Metabolic, mechanical and electrical properties of three motor-unit types.

Motor Unit Type	Metabolic Properties	Mechanical Properties	Electrical Properties
Type I (S-type)	Oxidative Low-myosin ATP'ase activity High-oxidative enzyme capacity Large number of mitochondria High myoglobin content Dark-red colored	Slow twitch Small force generation Small fiber diameter Most fatigue resistant Smaller motor unit size	Lower NCV Small axons Recruited at low force levels
Type IIa (FR-type)	Oxidative-glycolytic High-myosin ATP'ase activity Low-oxidative enzyme capacity Few number of mitochondria Low myoglobin content Pale-colored	Fast-twitch Large force generation Intermediate fiber diameter Fatigue resistant Intermediate motor unit size	Moderate NCV Intermediate axons Recruited at moderate forces
Type IIb (FF-type)	Glycolytic High-myosin ATP'ase activity Low-oxidative enzyme capacity Sparse mitochondria Low myoglobin content Pale-colored	Fast-twitch Large force generation Large fiber diameter Fatigable Large motor unit size	Higher NCV Large axons Recruited at high forces

2.4.2 Force Modulation:

Conversion of the electrical and biochemical energy to the mechanical energy in the skeletal muscle results in work done by the skeletal muscle. Thus, force is generated by this skeletal muscle in order to do work in achieving a certain task. The force is modulated by Motor Unit Recruitment and by the Motor Unit Activation Frequency or Firing Frequency which is also referred as Rate Coding [1, 2, 20, 26].

2.4.2.1 Motor Unit Recruitment:

Once a motor neuron is stimulated a motor unit is activated and generates a Motor Unit Action Potential (MUAP). MUAP is a compound potential representing the sum of the individual action potentials generated by the fibers and propagating along their sarcolemma and along their T-tubules in order to initiate the contraction of the muscle fibers belonging to that Motor Unit [7, 18].

The activation of one motor neuron will result in a weak but distributed muscle contraction. Therefore, more than one motor unit activation is needed to establish a significant contraction. The process where additional motor units are contributed to generate a muscle contraction at certain level is referred as motor unit recruitment [2, 24, 26]. Motor unit recruitment is the progressive activation of a skeletal muscle by successive recruitment of motor units to accomplish increasing gradations of contractile strength [26].

The relationship between the magnitude of the generated force and the motor unit recruitment is explained by Henneman's Size Principle [1, 2, 22]. In muscle contractions, motor units are recruited in an order from the weakest to the strongest in a characteristic sequence to produce contractions of increasing strength [19, 20]. According to the Henneman's Size Principle, this recruitment generally takes place in an orderly sequence being based on the size of the motor unit as contraction increases [19, 25]. The small motor units are recruited first, larger ones last. [2, 25]. Therefore, when a force of lower strength is required from a muscle innervated by more than one type of motor unit, this force is ensured by the slow twitch (S-type/Type I) motor units. As the strength of the force generated by the contraction increases, the fatigue-resistant (FR-type/Type IIa) motor units contribute to the contraction and then the fast-fatigable (FF-type/Type IIb) motor units are recruited in precise order according to the magnitude of the force produced by each unit [19].

Size-ordered recruitment serves two important purposes. First, it minimizes the development of fatigue by allowing the most fatigue-resistant muscle fibers to be used most of the time and by holding the more fatigable fibers in reserve until needed to achieve higher forces. Second, this recruitment ensures that the increment of force generated by successively activated motor units will be proportional to the level of force at which each individual unit is recruited. This provides to perform fine motor tasks [19].

2.4.2.2 Motor Unit Activation Frequency (Rate Coding):

The second way by which the force of a contraction can be modulated is by changing the firing rates of motor unit activation frequency of active motor neurons. This is a mechanism referred as rate coding [32]. When a muscle contraction is established as minimal as possible, the first motor unit recruited begins to fire irregularly at 2-3 Hz and then achieves a stable and fairly regular firing rate at 5-7 Hz. This is the onset frequency.

When the force of contraction is minimally increased, the first unit increases the rate of firing to 6-10 Hz. With further increase of muscle contraction, the second unit is recruited once the first unit achieves a firing rate of about 10 Hz. Slow units operate at a lower frequency range than faster units. Within that range, the force generated by a motor unit increases with increasing firing frequency [2].

2.4.3 Motor Unit Action Potential:

Muscle fibers generate action potentials individually when they are stimulated by the action potential conveyed through the motor neurons during a very weak contraction. These potentials are called “Single Fiber Action Potential (SFAP)”. The electrical activity recorded via the needle electrode is a compound potential belonging to a motor unit. This compound potential is the summation of these triphasic SFAPs and is referred as “Motor Unit Action Potential (MUAP)”. It has some parameters that are to be considered in the differential diagnosis of the neuromuscular diseases such as amplitude, duration and number of phase [6, 33, 34]. A MUAP is a signal of varying voltage as a function of time. It is illustrated with its electrophysiological characteristics in Figure 2.21.

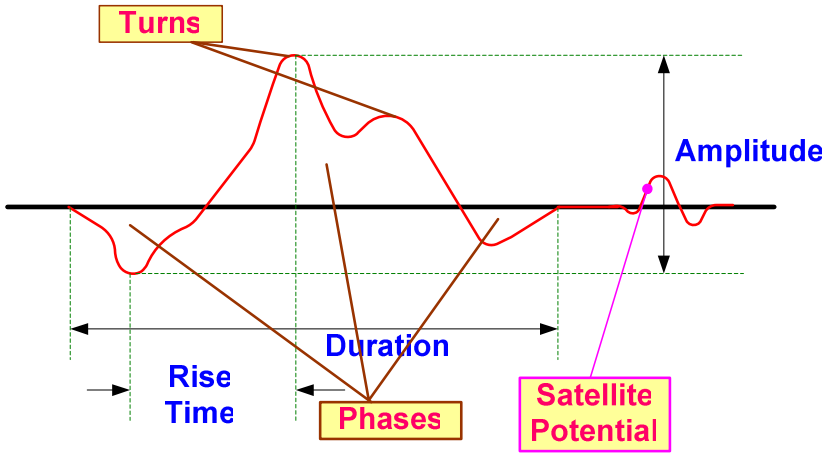

Figure 2.21. MUAP Parameters

Amplitude: The amplitude of a MUAP is a voltage value measured from peak to peak. It reflects the number of active muscle fibers of a motor unit within the uptake area of the electrode and the degree of synchronicity of firing of these muscle fibers. The peak-to-peak amplitude of the MUAP varies from 300 μV to 3 mV in normal subjects [3, 33, 34]

Duration: The duration is the time measured between the onset of the slow initial phase of the MUAP and the end of the slow terminal phase. It reflects the number of muscle fibers detected. The duration of a normal MUAP ranges between 3 and 15 milliseconds [3, 6, 33].

Phases: The phase is defined as that portion of a waveform between the departure from and return to the baseline. The number of phases is determined by counting negative and positive peaks. Normally, motor unit potentials have four or fewer phases [6, 33]. Number of phases indicates the complexity and the misalignment of the muscle fibers within a motor unit territory. In neurogenic diseases, polyphasic MUAPs arise due to slow conduction velocity in immature nerve sprouts or slow conduction velocity in reinnervated but still atrophied muscle fibers. In myopathic diseases, variation in muscle fiber size also causes polyphasic MUAPs [3].

Voltage Turns: These are the serrated potentials showing directional changes without crossing the baseline. They indicate desynchronization among discharging muscle fibers [3, 6, 33].

Rise Time: It is measured as a time lag from the initial positive peak to the subsequent negative peak. It helps estimate the distance between the recording tip of the electrode and the discharging motor unit. Since the resistance and the capacitance of the intervening tissue act as a high-frequency filter, a distant unit has a greater rise time. In general a rise time less than 500 μs ensures recording from within the motor unit territory [6, 33].

3. ELECTROMYOGRAPHY

3.1 General Principles of EMG

Since different muscles act on the same skeletal segment it is difficult to insert force sensors in series with the tendons. Thus, functional properties of muscles (i.e. their contractile properties) cannot easily be investigated in vivo. Besides mechanical properties, the activity of skeletal muscles is also associated with the generation of electric signals that can be recorded by electrodes inserted in the muscle (intramuscular recordings) or fixed over the skin (surface recordings). The electric signals generated by the muscles during their activity are referred to as electromyographic (EMG) signals [1].

Electromyography refers to the process of studying and recording the electrical activity of muscle (i.e. MUAPs) [3, 10]. The electrical activities recorded by EMG are insertional potential, normal and abnormal spontaneous activities such as miniature end-plate potential (MEPP), fibrillation potentials, positive sharp waves, complex repetitive discharges, fasciculations and myotonia, activities of the voluntary contractions such as motor unit action potential (MUAP), recruitment pattern and interference pattern. The frequency range of MUAPs recorded during electromyography is between 10 Hz to 10 kilohertz [42]. Their amplitudes range from 200 μV to 3 mV and have a duration of 3 to 15 milliseconds when they are recorded by a concentric needle electrode [6, 10, 35].

EMG has been used as an auxiliary electrodiagnostic method in the diagnosis of neuromuscular disorders. It has been used to detect and characterize the disease processes affecting the motor units [10]. It is beneficial in the extension of the clinical evaluation when it is difficult to find the localization the pathology from where the signs and symptoms originate [6, 10, 26, 36]. The findings allow the clinicians to localize the

lesions to the anterior horn cell, nerve roots, peripheral nerves, neuromuscular junctions or muscle fibers of the motor unit in question [6, 10].

As a result, EMG ensures an electrophysiological technique helpful to clinicians in differentiating neurogenic, myopathic and neuromuscular junction disorders from each other.

3.2 General Structure of EMG instrument

The biopotentials acquired from the muscles via the needle electrodes are usually in the levels of hundreds of microvolts to several millivolts. They should be amplified to perceivable voltage levels to be evaluated by the clinicians. Electrode constitutes the passive electrical interface between the patient and the EMG system. Active electronic circuitry where the electrode is directly attached is referred to as the input stage or the front end of the EMG amplifier [1]. In addition these biopotentials should be filtered in order to eliminate the unwanted noise.

Modern EMG instruments comprise amplifier stages, filters, analog-to-digital converters (ADC), a computer system with a specific software that are used to process the acquired signals for different electrophysiological applications and that store the processed data, a loud speaker fed by an audio amplifier, a display to monitor the acquired signal and a stimulator to be used in nerve conduction studies in conjunction with EMG system [1, 3]. The general structure of an EMG instrument is summarized in Figure 3.1.

3.2.1 Sources of Interferences:

Sources of interference such as power line cables, externally and internally generated magnetic and electrical fields, HF interference and ripples in the power supply of the amplifier can degrade the quality of the signal [1]. The block diagram of the relationship between these interferences and the patient and the EMG instrument is represented in Figure 3.2.

A primary source of interference is due to capacitive coupling of the measurement cables with power lines (C_{c1}, C_{c2}); the induced currents (i_{c1} and i_{c2}) flow through the body via the electrodes and close to earth by means of the body-earth capacitance (C_{body}) and the isolation capacity (C_{iso}) in series with the reference contact impedance (Z_{ref}).To reduce this effect, the parasitic capacitances C_{c1} and C_{c2} should be kept as low as possible. This can be achieved by making the connection between the front-end and the electrodes as short as possible [1, 37].

One of the other interference sources originates from the common mode voltage, V_c generated by the capacitances between the body and the power line [1].

Another source of interference comes from high-frequency (HF) emissions of neighboring equipment located in the vicinity of the EMG instrument. Even though there is a greatly reduced CMRR, the effects of the HF itself in the signal are the same as in the case of power line interference. However, the presence of a low-pass filter in the signal conditioning solves this problem [1].

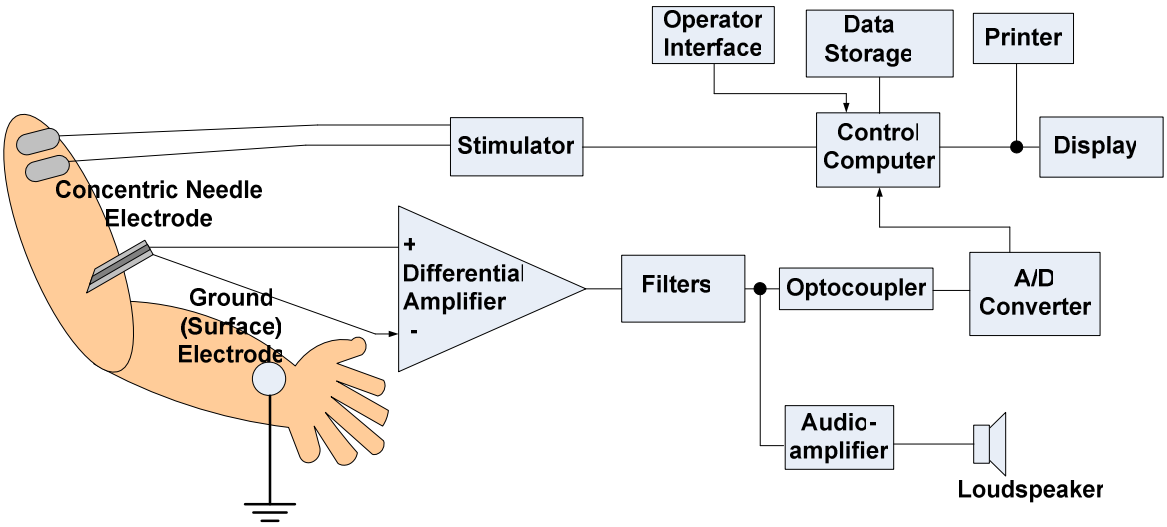

Figure 3.1 Basic Components of the EMG Instrument.

3.2.2 The Amplifier:

An amplifier takes this small signal voltage and increases its amplitude so that it can be processed and displayed. The high-input impedance amplifiers alters the weak low voltage signals into stronger, higher voltage signals eligible for analog to digital (A/D) conversion [38]. On the other hand, these EMG signals should be amplified without magnifying the interference signals [26, 35]. The interference signals originating not only from the power lines but also distant muscle action potentials are referred as common mode voltages [26]. A differential amplifier system is used in EMG instruments to prevent the amplification of these signals. While using a concentric needle electrode (CNE), there are two inputs coming from either the active electrode of CNE or the reference electrode of CNE. These inputs are the voltages measured with respect to a third ground electrode. Differential amplifier amplifies only the voltage difference between two input channels connected to the recording electrodes. The common mode voltages taking place between two input terminals where the recording electrodes are connected and common ground are rejected by this differential amplifier. The ability of

rejecting common mode signals of a differential amplifier is specified by its common mode rejection ratio (CMRR). Theoretically, at the CMRR is higher, better rejection is obtained. Good differential amplifiers have usually CMRR of almost 100,000 or 100 dB on logarithmic scale [10, 26, 35].

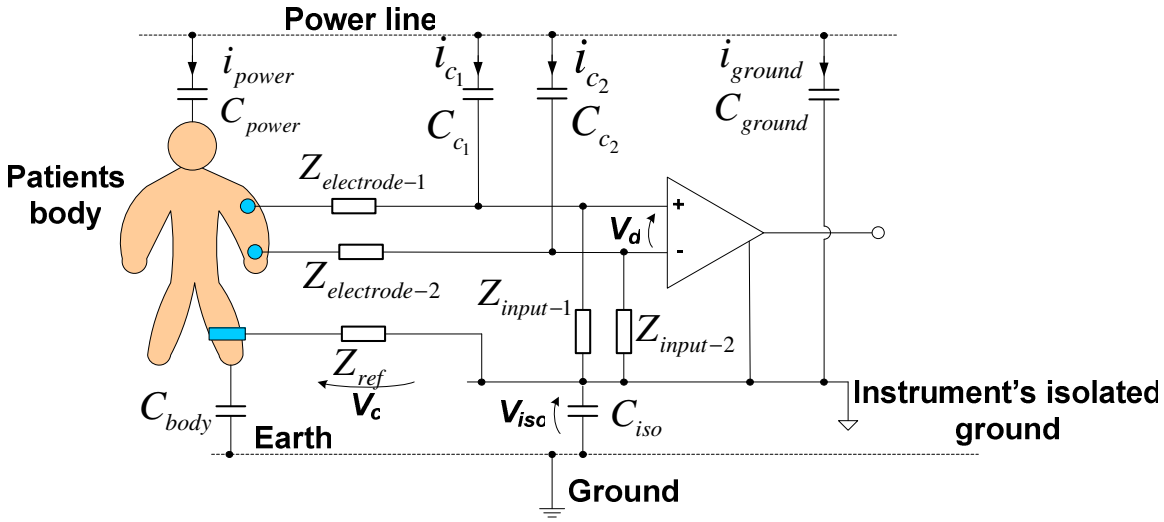

Figure 3.2 Working environment of an EMG instrument; the body of the patient and the instrumentation itself are connected to power line cables and earth through parasitic capacitances. C_{power} = parasitic capacitance between body and power line; C_{body} = capacitance between body and earth; C_{c1}, C_{c2}= parasitic capacitances between power line and input cables; C_{iso}= parasitic capacitance of the isolation barrier; C_{ground} = parasitic capacitance of the power supply between power line and isolated ground; $Z_{electrode-1}$, $Z_{electrode-2}$= electrode contact impedances; $Z_{input-1}$,$Z_{input-2}$= amplifier input impedance; Z_{ref} = patient reference contact impedance; V_c= common mode input voltage; V_d = Differential input; V_{iso}= isolation mode voltage.

Another parameter that should be considered in biopotential amplifiers in EMG instruments is the input impedance. While recording muscle action potentials the tissue impedances and that of the electrode wires can be considered as negligible compared with

those at the needle and at the input terminal of the amplifier. They act as a voltage divider with voltage changes in proportion to the respective impedances [10][26][35]. Higher input impedances of the amplifier are required to prevent the attenuation of the signal. It also ensures the improvement of the CMRR by reducing the electrical imbalance of the recording electrodes [1, 10, 26]. The input impedance of such an amplifier should be at least 10 MΩ [10, 35].

3.2.3 Filters:

In order to reduce artifacts and noise accompanying the EMG signals, EMG instruments possess low-pass and high-pass filters. Generally, the frequency band provided by these filters ranges between 2 Hz and 10 kHz in the clinical studies [26, 35, 39].

The high-pass filtering is needed to remove the unwanted DC offset generated by the half-cell potential of the electrodes, which is DC-coupled in the input stage and, in lower percentage, by the offset voltage of the input amplifier itself. If this offset is amplified together with the EMG signals by the next amplification stages, it could reduce the useful dynamic range of the signal or even saturate the last stages [1].

Low-pass filtering is required to remove unwanted noise beyond the bandwidth of interest that could cause a degradation of the signal-to-noise ratio (i.e., the ratio between the amplitude of the signal and the background noise) [1].

In the modern EMG instruments, the cut-off frequencies can be adjusted for various EMG techniques. As an example, the high-pass filter is set to 500 Hz in single fiber EMG

(SFEMG). Therefore, the volume-conducted interfering activity from distant muscle fibers is minimized [26, 40].

In surface EMG amplifiers, there are also notch-filters to reduce common-mode interference yielding from the 50 or 60-Hz power lines being located closer to the measurement site [1, 38].

3.2.4 Galvanic Isolation

According to the safety regulations in medical care, patients should be isolated from the non-biomedical equipment that can be connected to an EMG amplifier. No resistive conductive path should exist between the EMG amplifier and any external equipment that can be connected to it. This can be ensured by galvanic isolation. This condition can be obtained by optical coupling the circuits connected to the patient (also called the patient circuit), while supplying the power by means of a medical grade, transformer-isolated power supply (DC/DC converter) or a battery [1, 40].

The optocoupling can be either analog or digital. Analog optocoupler are intrinsically noisy and should be placed at the end of the amplification chain. Digital coupling can be implemented after A/D conversion, and generally allows better performance both in terms of noise and lower value of the isolation capacitance [1, 41]. A low value of isolation capacitance is required not only as a safety requirement, but also because it is a potential source of interference on the input signal [1]. The galvanic isolation after the input stage and the filter stage is shown in Figure 3.3.

3.2.5 Analog-to-Digital (A/D) Converters:

After the optoisolation stage, the EMG signal is usually sent to an acquisition system for data recording and/or online display. The acquisition system can be a series of A/D (analog-to-digital) converters, residing internally to the EMG instrument as in case of portable EMG acquisition system, such as EMG data loggers or an external PC acquisition board [1].

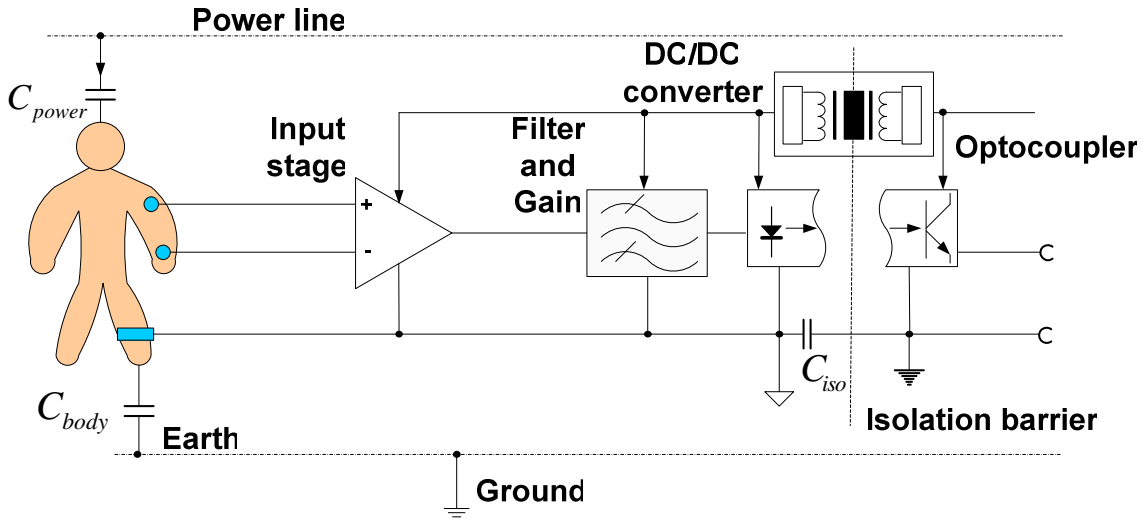

Figure 3.3 Block diagram of an EMG amplifier, showing the power supply rails, the galvanic isolation circuitry, and its parasitic isolation capacitance, C_{iso}.

New generation EMG equipments comprise computers and software where the acquired signals are processed, displayed and stored. The EMG signals picked up by the electrodes are analog signals. They should be converted into digital format to be handled by the computer system of the EMG system. This is achieved by analog-to-digital converters (ADC) built in these instruments.

Before being fed into A/D converter, analog signals must be sampled. Sampling is the process of acquiring the values of the signal at equally spaced time intervals, which define the sampling frequency of the system [1].

Beside the advantages of the digital format, the digitization results in the modification of the original signal. The fidelity of the digitized EMG signal depends on the resolution of the system. The time resolution which implies the sampling rate (SR) over a sweep can be up to 100 kilo samples per second. The amplitude resolution of the recent EMG systems ranges from 12 to 16 bits [38].

3.2.6 Display:

The waveform of the MUAPs picked up by the EMG electrodes can be monitored by means of a visual display. In the older generations of EMG systems, cathode ray tube (CRT) oscilloscopes were used for this purpose. The implementation of the digital technology into the recent EMG systems has enabled to use LCD displays.

While monitoring the EMG signal on the display, it is possible to follow and adjust the sensitivity that is usually expressed as amplitude value in microvolts or millivolts per division in centimeters [6]. In commercially available EMG systems, the sensitivity ranges from several microvolts per division to 20 mV per division.

3.2.7 Loudspeaker:

MUAPs have distinct auditory characteristics for different clinical states. These potential are fed to an audio-amplifier and then to loudspeaker [6, 42]. While following visually the waveform of these signals from the display the clinicians usually assess the characteristic sounds of various EMG patterns belonging to different clinical cases [10, 26].

3.2.8 Stimulator:

Nerve conduction studies are usually performed in conjunction with the EMG and EMG instruments contain a stimulator for this purpose [43]. An electrical stimulus with varying intensity is generated and is applied to the nerve being studied through the surface electrodes. Then, the response of the nerve is acquired with another electrode. Motor nerve conduction studies, sensory nerve conduction studies and mixed nerve conduction studies are performed with the stimulator of the EMG systems [1, 42].

3.3 Electrodes

In order to display the EMG signals on an EMG system the MUAPs generated by the muscle fibers during the voluntary contraction should be picked be through electrodes. There are kinds of electrodes of recording electrodes: needle electrodes and surface electrodes. There are also different types of needle electrodes. These are concentric needle electrode (CNE), monopolar electrode, single fiber electrode (SFE), macro-electrode.

3.3.1 Concentric Needle Electrode (CNE):

The concentric needle electrode consists of a platinum wire located centrally inside a hollow needle being completely insulated from this hollow needle. This platinum wire is the active electrode with a 150 µm diameter [30, 36]. The outside cannula is the reference electrode [44]. The outside diameter of the needle is approximately 0.3 mm [6]. The tip of the cannula is beveled to a 15 ° angle yielding an elliptical recording surface area.

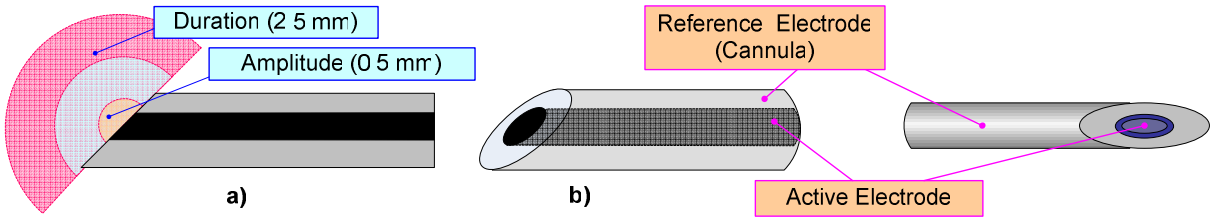

Figure 3.4 a) Uptake area of the Concentric Needle Electrode **b)** Structure of the CNE

The dimensions of this surface are 150×580 µm. This surface are is 0.07 mm^2. The CNE has an impedance of 50 kΩ [26, 30, 38]. The CNE electrode is illustrated schematically in Figure 3.4.

The wire and the shaft constitute a hemispheric recording territory at the tip of the electrode. When the CNE is inserted in the vicinity of a source of electrical activity it registers the potential difference between the wire and the shaft. This is achieved in a restricted recording area. A surface electrode serves as a ground [26, 38].

The MUAP amplitude recorded with this electrode is directly related to the number and size of muscle fibers located within a hemisphere with a radius of 0.5 mm at the needle tip. The MUAP duration is related to the fibers located within a semiglobe with a radius of 2.5 mm [6, 26, 38, 45-47].

3.3.2 Monopolar Needle Electrode (MNE):

The monopolar needle electrode (MNE) is an insulated needle with a conical recording surface [30, 38]. It has a fine point and is made of stainless steel [6][26]. The insulation of the MNE is achieved by coating the needle with Teflon^{TM} except at the distal 0.2 to 0.4 mm [6, 26, 38].

MNE has a recording surface with an area of 0.15 to 0.25 mm^2 [30, 38]. The average diameter of wire and the Teflon sleeve of the MNE is 0.8 mm [6, 26]. The length of the MNE varies from 12 mm to 75 mm [6]. The average impedance lies between the ranges from 1.4 MΩ at 10 Hz to 6.6 kΩ at 10 kHz [26].

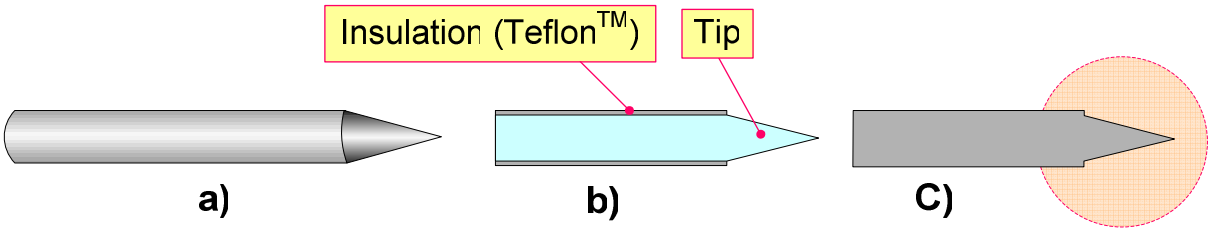

Figure 3.5 a) Monopolar Needle Electrode (MNE). **b)** Longitudinal sectional representation of the MNE. **c)** Up-take area of the MNE.

For needle EMG studied MNE is used as the active electrode. A surface electrode or a second needle in the subcutaneous tissue is used as a reference electrode. A separate surface electrode placed on the skin is used as the ground electrode [6, 26, 38, 44].

MNE is less painful, less expensive and more sensitive in identifying fibrillation and positive sharp waves than the concentric needle electrode [6]. However, it is less stable and hence noisier than CNE [26]. It also records activity from more distant MUs giving a more unstable baseline, which renders this type of recording somewhat more difficult for certain types of automatic analysis [30].

Since it is not a directional electrode, the spatial recording characteristics of the MNE differ from those of the CNE. It can detect more muscle fibers in its neighborhood of its active recording area resulting in the MUAPs tending to be more polyphasic and to have longer duration and higher amplitude [38, 46]. It registers a MUAP being almost twice in amplitude and more complex in shape [26].

MNE is sometimes used as a recording electrode to record compound muscle action potential (CMAP) from the belly of the muscle in repetitive nerve stimulations. It can be used as a stimulating electrode when the nerve is deeply located. It can also be used for intramuscular stimulation when only a single or a few motor units need to be activated at a time [6, 38]. MNE is illustrated in Figure 3.5.

3.3.3 Single Fiber EMG (SFEMG) Needle Electrode:

In order to study electrical activity from a single muscle fiber, selective recording is required [39]. Since the leading-off surface of 150 ×580 μm for CNE is large compared

to the diameter of a muscle fiber of 25 µm to 100 µm, specially designed and manufactured electrodes that have smaller leading edge are needed for this purpose are needed [6, 26, 40].

The SFEMG electrode comprises a platinum wire with a 25-µm diameter inside a cannula used as an active electrode. This small recording surface provides selectivity which is necessary in recording action potentials from individual muscle fibers [48]. The tip of this wire is bent toward the side of the cannula opposite to the beveled surface to avoid recording from mechanically compressed or damaged muscle fibers [26, 38, 39, 49]. The surface of this wire is located just a few millimeters behind the tip [26, 39]. SFEMG electrode is iluustrated in Figure 3.6.

The wire is insulated by an epoxy resin throughout the entire cannula. The diameter of the cannula is 0.5 to 0.6 mm. The cannula is used as a reference electrode and is connected to the amplifier input. The patient is grounded with a surface electrode [39].

SFEMG electrode has an uptake radius of about 300 µm [49]. This allows to record action potentials of single muscle fibers of relatively large amplitude such as 1 to 6 mV [39].

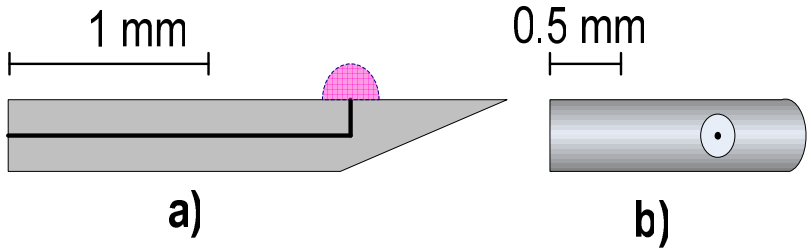

Figure 3.6 SFEMG electrode: **a)** SFEMG electrode with its uptake area **b)** active electrode surface on the side port.

The bioelectric sources located of this uptake area are perceived as the common-mode signals and suppressed by the differential amplifier [3].

3.3.4 Macro EMG Electrode:

Macro EMG needle electrode consists of a modified SFEMG electrode and a steel cannula [38]. The SFEMG electrode is 25-µm platinum wire exposed in a side port and is used to trigger the record using an action potential from a nearby muscle fiber. The cannula has a 0.55-mm diameter and a 15-mm exposed surface used to record the electrical activity of the entire motor unit [6, 30].

Concentric macro or ConMac electrode was developed as a modification of the macro EMG electrode. ConMac electrode uses a modified CNE which is coated with Teflon™ to within 15 mm of its tip to trigger an action potential nearby muscle fibers [38]. The macro EMG electrode and the ConMac electrode are represented in Figure 3.7.

A surface electrode placed on the skin is used as a reference electrode with the Macro EMG electrode during the recording process [30].

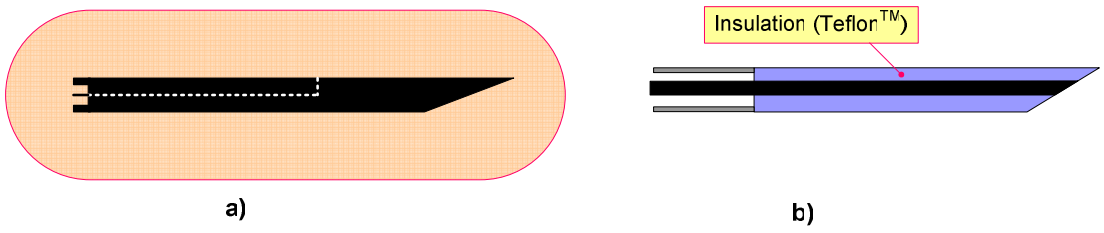

Figure 3.7 a) The macro EMG electrode with its uptake area b) ConMac electrode.

3.3.5 Surface Electrodes:

A surface electrode consists of a conductive medium with defined dimensions and shape, which is electrically connected to the skin of the patient and are round or square metal plates. These electrodes are manufactured materials such as solid silver or gold, platinum, sintered silver and silver chloride, carbon, and sponge saturated with electrolyte gel or conductive hydrogel. These electrodes have an average dimension of 1×1 cm. They are usually affixed to the skin with an adhesive tape. An electrolyte cream is applied under the electrode in order to reduce the impedance of the skin-electrode interface [1, 6, 26].

Surface electrodes have a large pick-up area. They register electrical activity from a wider region with a 20-mm diameter. They are non-selective. However, they record mass activity from a large proportion of a muscle, or from many muscles, depending on recording site [26, 30].

They are suitable for monitoring voluntary muscle contraction during kinesiologic studies and evoked compound nerve and muscle potentials. They are commonly used polygraphic kinesiological recordings, usually as simultaneous recordings from many muscles in the mapping of muscle activation pattern in gait analysis, ergonomic studies, rehabilitation studies, sports medicine. It may also be used to map abnormal muscle activation pattern in movement disorders, especially in cervical dystonia or writers' cramp before the treatment with botulinum toxin [26, 30].

The surface electrode also serves as a stimulating probe, a reference and a ground lead in conjunction with monopolar needle electrode or with concentric needle electrode.

Since they fail to reproduce high-frequency components adequately, they cannot be used as an active electrode in studying MUAPs [26].

The structure of a surface electrode and it s uptake area compared with motor unit having a 10-mm diameter is represented in Figure 3.8.

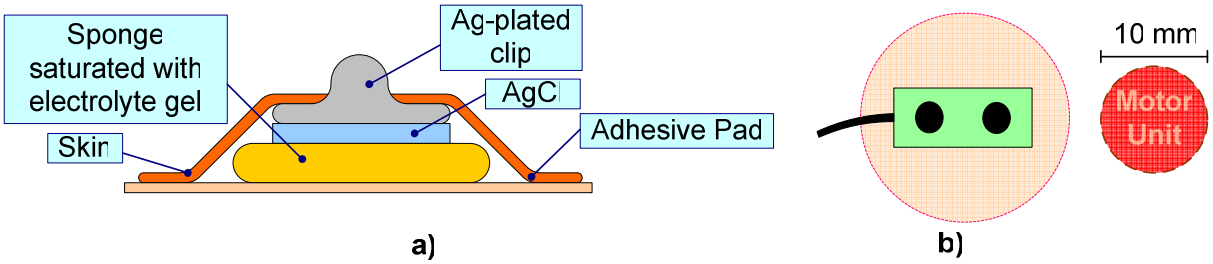

Figure 3.8 Surface EMG electrode **a)** Cross-section of a surface EMG electrode;**b)** Plate-mounted surface electrode and its uptake area compared to motor unit.

3.4 EMG Signals in Clinical Studies

3.4.1 Insertional Activity:

Insertion of a needle electrode into the muscle or quick movement of the inserted needle electrode through muscle causes the depolarization of the muscle fibers. This activity lasts on average several milliseconds [24, 26]. The waveform appears as positive or negative high-frequency spikes in a cluster as shown in Figure 3.9.

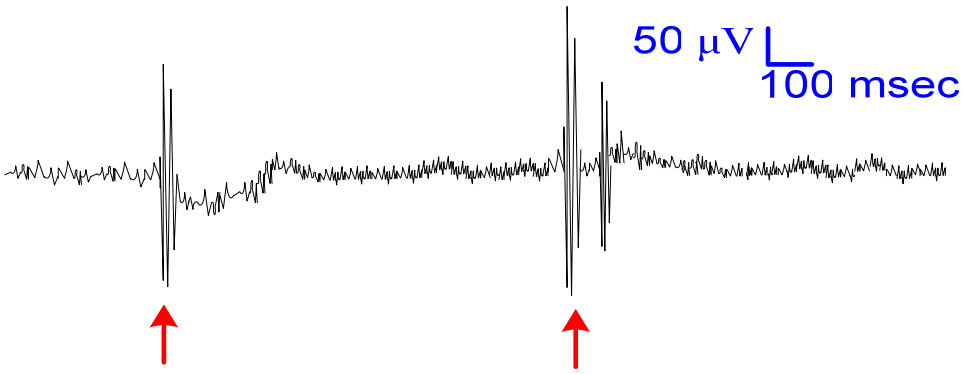

Figure 3.9. Insertional activity occurring with needle movements indicated by arrows.

Since this activity originates from muscle fibers injured or mechanically stimulated, it is also referred as injury potential [26].

The insertional activity can be helpful to confirm that the needle electrode is in muscle rather than fat or subcutaneous tissue [24].

3.4.2 Spontaneous Activity:

3.4.2.1 Analysis of Spontaneous Activity:

The recognition of the abnormal spontaneous activity is one of the important aspects of the needle EMG examinations in clinical studies. The practical information provided by the presence of the abnormal spontaneous activities can be categorized as follows;

i) Distribution of the spontaneous activity: It can help to estimate the neuroanatomic localization of the lesion.

ii) Types of spontaneous activity: Since certain types of spontaneous activities are associated only with specific disorders it can provide specific diagnostic information.

iii) Degree or amount of spontaneous activity: It often helps to determine the severity of the disease.

iv) Time course of the disease: For example, in a radiculopathy, several weeks must be spent before fibrillation potentials are observed in the limbs.

The waveforms observed by means of the needle EMG should be systematically analyzed in terms of morphology, stability and firing characteristics.

The source of the spontaneous activity can be identified by its morphology including the size of the potential (amplitude, duration, number of phases) and its initial deflection. By defining the source generator, the type of the spontaneous discharge can be identified. The source generators are as follows: (1) Neuromuscular junction (NMJ), (2) a single muscle fiber, (3) the terminal axon twig, (4) a motor neuron and/or axon, (5) multiple muscle fibers linked together. These source generators are illustrated in Figure 3.10.

The spontaneous activity occurring at the NMJ is the miniature end-plate potentials (MEPPs) which are also called end-plate noise. They have small amplitudes and monophasic negative morphology [24].

In a single muscle fiber, the spontaneous activity referred as muscle fiber action potential (MFAP) occurs when the muscle fiber depolarizes to threshold. There are two basic morphologies for this activity such as brief spike and positive wave. The brief spike has duration varying between 1 to 5 milliseconds with low amplitude (typically 10-100μV) and can be biphasic or triphasic. It is seen when a muscle fiber depolarizes

spontaneously. On the other hand, positive wave is has a positive phase followed by long negative phase. Denervating potentials such as positive sharp waves and fibrillation potentials are examples for MFAPs [24].

The spontaneous discharges arising from the motor neuron or from their axons can cause a potential with the morphology of a motor unit referred as motor unit action potential (MUAP). Fasciculations, tetany, myokymic discharges, neuromyotmic discharges and cramps are the abnormal spontaneous MUAPs [24].

The spontaneous discharges generated by the time-linked muscle fibers are the multiple different muscle fiber action potentials. They exhibit consecutively fired and distinguishable spikes that are linked together over the time axis. Complex repetitive discharges constitute important example to this category of spontaneous activity [24]. The morphologies of the spontaneous activities are summarized in Figure 3.11.

Beside the morphology of the spontaneous waveform, the stability is also important in the assessment of this activity. If the morphology of the potential changes, the decrements or changes abruptly should be noticed. MFAPs that wax and wane are observed in myotonic discharges. Amplitude of the MUAP is decremented markedly in neuromyotonic discharges [24].

Another factor in the assessment of the spontaneous activity is the firing characteristics that imply regularity and firing rate. For example, complex repetitive discharges are perfectly regular. If a spontaneous discharge is irregular, it can be sputtering as in case of end-plate spikes, waxing/waning as seen in myotonic discharges, or waning like neuromyotonic discharges. Bursting pattern which means the relative

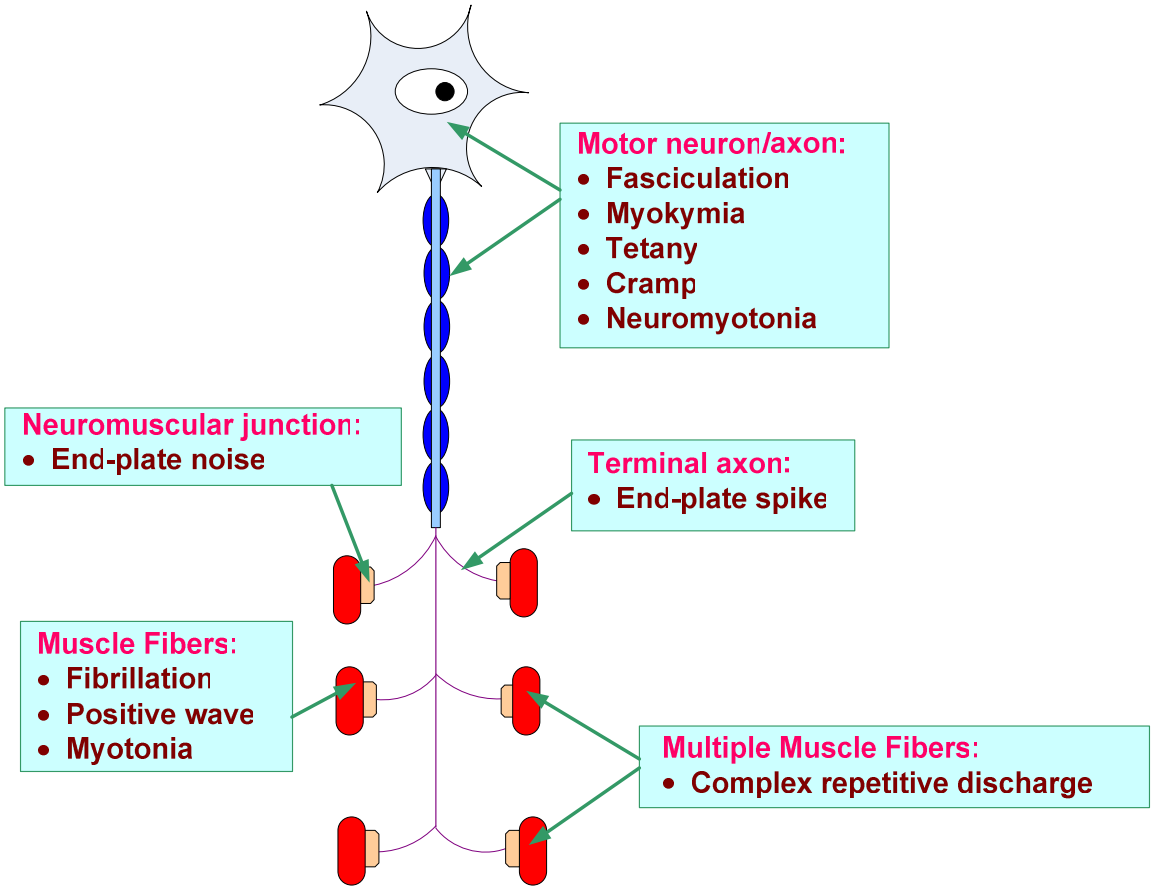

Figure 3.10. Schematic representation of the source generators of the spontaneous discharges

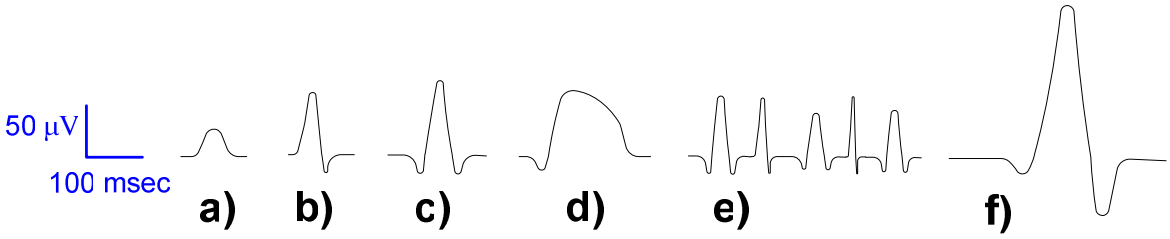

Figure 3.11. Morphologies of the spontaneous waveforms: **a)** miniature end-plate potential, **b)** muscle fiber action potential (biphasic), **c)** muscle fiber action potential (triphasic), **d)** muscle fiber action potential (positive wave), **e)** multiple muscle fiber potentials linked together, **f)** motor unit action potential.

electrical silence between groups of discharges can exist. This is observed as a characteristic pattern of tetany and myokymic discharges. Slow firing rates are typical for spontaneous activities. Firing rates less than 4-5 Hz indicate that the discharges cannot be voluntary. On the other hand, very high firing rates such as 150 to 250 Hz are characteristic for cramps or neuromyotonic discharges [24].

3.4.2.2 Normal Spontaneous Activity

3.4.2.2.1 End-Plate Noise:

It yields from the normal spontaneous exocytosis of individual quanta of acetylcholine (ACh) diffusing across the NMJ causing a sub-threshold end-plate potential that do not propagate along the muscle fiber [24, 26]. It is an irregular monophasic negative potential being 10-50 μV in amplitude and 1-2 milliseconds in duration [26]. These potentials that fire irregularly at 20 to 40 kHz represent physiologically miniature end-plate potentials (MEPPs). They can be recognized by means of seashell sound heard on the loudspeaker of the EMG instrument by the examiner in clinical practice [24, 26]. The waveform of the end-plate noise is illustrated in Figure 3.12.

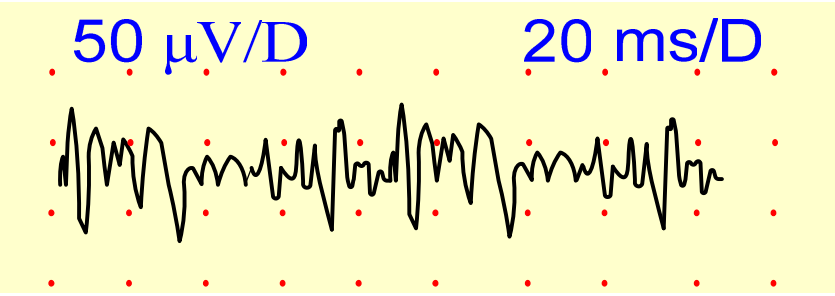

Figure 3.12. End-plate noise.

3.4.2.2.2 End-plate Spikes:

End-plate spikes are the MFAPs resulting from discharges of single muscle fibers excited by the needle. These are intermittent spikes being 100-200 μV in amplitude and 3-4 milliseconds in duration are fire irregularly at 5-50 Hz. They are biphasic with an initial negative deflection. The initial negativity of the waveform indicates that the needle electrode is at the site where the action potential is generated. They are suggested to occur as a result of a needle-induced irritation of a terminal nerve twig. End-plate spikes are differentiated from the fibrillation potentials which are also brief spikes through their initial negative deflection and their highly irregular firing rate [24, 26]. The end-plate spikes are shown in Figure 3.13.

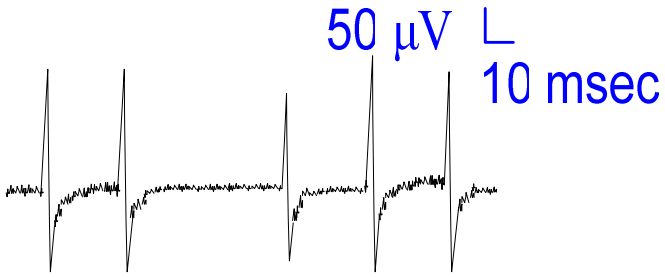

Figure 3.13 End-plate spikes.

3.4.2.3 Abnormal Spontaneous Activity:

Muscle at rest is normally silent outside of the end-plate zone. Any persistent spontaneous activity that lasts longer than 3 seconds is abnormal. Spontaneous activity may take place when the needle is placed in the muscle or may be induced by needle movement, muscle contraction or electrical stimulation [24].

3.4.2.3.1 Abnormal Spontaneous Activity

3.4.2.3.1.1 Fibrillation Potentials

Fibrillation potentials are action potential arising spontaneously from single muscle fibers. They results in the spontaneous depolarization of the muscle fibers. They are the indicator of the active denervation. They are often associated with neurogenic disorders such as neuropathies, radiculopathies or motor neuron disease. However, they can be seen also in some myopathic disorders such as inflammatory myopathies and dystrophies and occasionally in neuromuscular junction (NMJ) disorders (e.g. botulism) [24, 36].

The waveform of this potential exhibits a brief spike with an initial positive deflection with a 1-5-millisecond duration and with 10-100-μV amplitude. Its firing pattern is regular with frequency 0.5 to 10 Hz [10, 24, 26]. The waveform of fibrillation potential is illustrated in Figure 3.14.

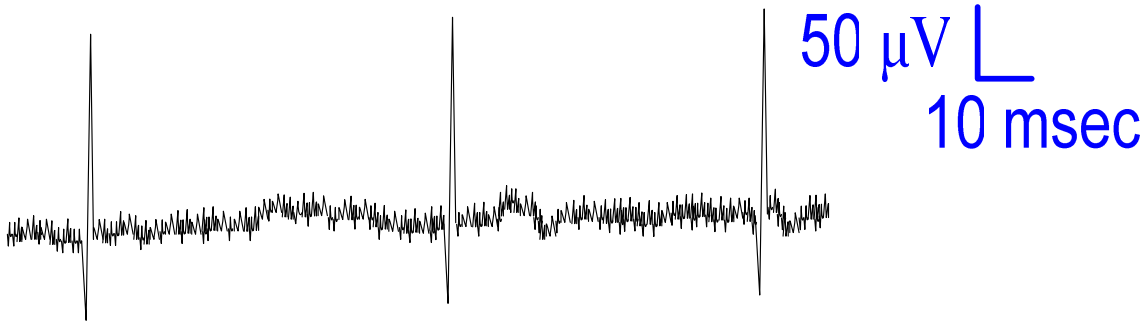

Figure 3.14 Fibrillation Potential

3.4.2.3.1.2 Positive Sharp Wave:

Positive sharp waves are the spontaneous activities resulting in the single-fiber activity like the fibrillation potentials. They indicate also active denervation [24].

They arise when the needle tip damages a fiber and spontaneous action potentials propagate up to the needle tip and then are damped. The waveform is determined by the physical relationship between the generator (i.e. muscle fiber) and the recording electrode. The muscle fiber is mechanically deformed by the needle electrode and that segment of the fiber becomes inexcitable. The depolarization occurring at the distal of the needle can propagate toward the needle and it results in a sharp positive discharge. As the propagation approaches to the needle a negative deflection which slowly extinguishes takes place [10, 24, 26, 36].

Positive sharp waves have almost the same clinical significance as the fibrillation potentials. They can be observed in many common disorders such as radiculopathy and entrapment neuropathies [24].

The waveform has a saw-tooth appearance with a brief initial positivity followed by a slow electro-negativity much lower in amplitude but longer in duration. They have variable amplitude such as 10 to 100 μV which can occasionally reach to 3 mV. The firing frequency of the positive sharp wave ranges between 0.5 to 10 Hz. The waveform for positive sharp wave is shown in Figure 3.15.

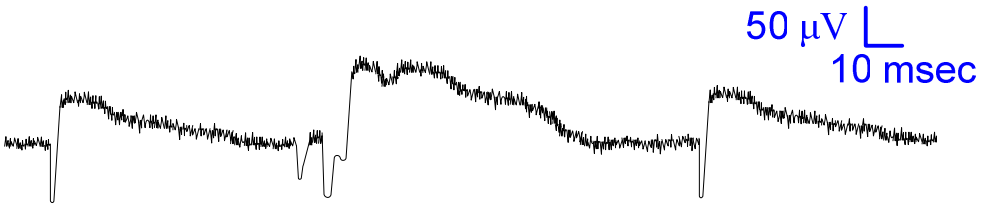

Figure 3.15 Positive Sharp Wave with a characteristic sharp positive discharge followed by slow negativity.

3.4.2.3.1.3 Complex Repetitive Discharge:

This is an spontaneous activity yielding from the depolarization of a single muscle fiber followed by the ephaptic transmission between adjacent fibers (i.e. direct spread from sarcolemma to sarcolemma) [24, 36].

The waveform is characterized by time-linked individual spikes firing consecutively. Complex repetitive discharges (CRD) range 50 μV to 1 mV in amplitude and 50 to 100 milliseconds in duration [26]. They have a constant frequency between 1-100 Hz [36]. On EMG, they are recognized as the high-frequency multiserrated repetitive discharges with an abrupt onset and termination [24, 36]. The pattern of the complex repetitive discharges is shown in Figure 3.16.

They are observed in myopathies such as muscular dystrophies and polymyositis and in chronic denervating conditions such as motor neuron disease, radiculopathy, chronic polyneuropathy and myxedema [26, 36].

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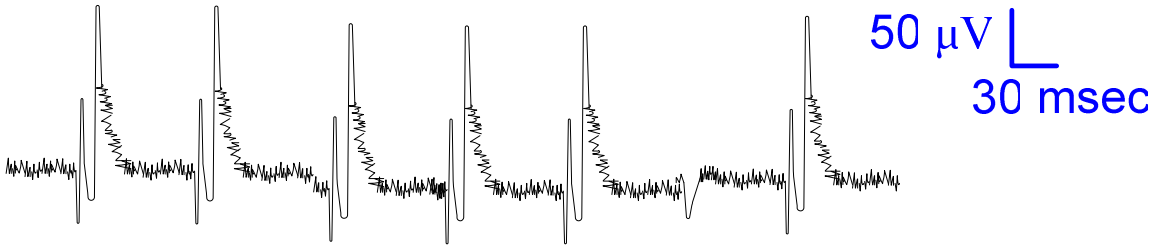

Figure 3.16 Complex Repetitive Discharge.

3.4.2.3.1.4 Myotonic Discharges:

A myotonic discharge is a spontaneous discharge generated by a muscle fiber like fibrillation potentials and positive sharp waves. However, it is differentiated by its characteristic waxing and waning of both amplitude and frequency. It has a firing rate of 20 to 150 Hz. Since the source generator is a muscle fiber, its waveform may be a positive wave or a brief spike [24]. The waveforms of the myotonic discharges are shown in Figure 3.17.

Myotonic discharges are typically seen in muscle fibre membrane channelopathies, including dystrophia myotonica, congenital myotonias, proximal myotonic dystrophy, and hypokalaemic periodic paralysis. They can be seen also in several forms of myopathies such as acid maltase deficiency, polymyositis, myotubular myopathy [24] [36].

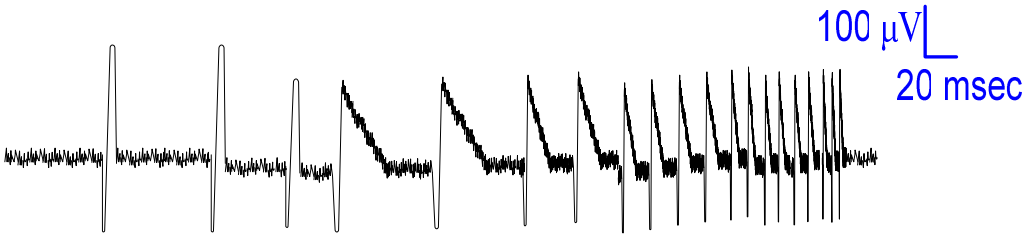

Figure 3.17 Myotonic Discharges

3.4.2.3.2 Abnormal Motor Unit Potentials:

3.4.2.3.2.1 Fasciculation Potentials

Fasciculation Potentials are the involuntary spontaneous discharges of a group of muscle fibers representing either a whole of an individual motor unit or part of a motor unit [24, 26, 36].

Fasciculations near the surface of a muscle may be visible at the skin. On the other hand, those occurring deep within the muscle and detected by an EMG needle are not visible [26, 36].

In contrast to voluntary motor unit potentials, fasciculation potentials may undergo slight changes in amplitude and have irregular complex waveforms representing reinnervated motor unit [24, 26]. They are isolated discharges that recur at irregular intervals, usually in the order of several seconds [36]. Fasciculation potentials usually fire very slowly and irregularly, less than 1-2 Hz. However, voluntary motor unit potentials

start to fire at 4 to 5 Hz when a patient is asked to slightly contract a muscle and cannot fire more than these frequencies [24]. The waveforms of the fasciculation potentials are represented in Figure 3.18.

Fasciculations probably arise within the fine terminal arborisation of a single motor axon within the muscle. It has been shown that there may be more than one generator of a single fasciculation as evidenced by subcomponents of the potential occurring in a different order on different discharges. Thus other source generator of fasciculation potentials is the motor neuron and its axon [24, 36]

In clinical examination, fasciculations are recognized by the brief twitches that occasionally yield from the apparent movement of a joint [24]. Fasciculations are associated with several lower motor neuron diseases such as amyotrophic lateral sclerosis. On the other hand, they can be seen in other neurogenic diseases such as radiculopathies, polyneuropathies and entrapment neuropathies, thyroid disease, and peripheral nerve hyperexcitability syndromes [24, 36]. Fasciculations may be observed transiently after the administration of the anticholinesterase medications or a depolarizing neuromuscılar blocker. These potentials remain for about four days after the removal of the nerve supply and then they disappear [26]. In addition, normal individuals may have some fasciculations referred as benign fasciculations. They fire faster when compared with malign fasciculations. Unlike the malign fasciculations seen pathologic conditions such as motor neuron disease benign fasciculations affect the same site repetitively (e.g. eyelid twitching) [24]. In contrary to malign fasciculations benign fasciculations are not accompanied by denervation changes and are not associated with muscle weakness, wasting or any abnormal reflexes [24, 36].

3.4.2.3.2.2 Doublets, Triplets and Multiplets

Spontaneous MUAPs that fire in groups of two, three or multiple potentials are called doublets, triplets and multiplets respectively [24, 26]. These are seen with fascucilation potentials and can be observed in any case where fasciculation potentials take place such as neurogenic conditions. However, they can be also characteristically seen in tetany from hypocalcemia [24]. Doublets are illustrated in Figure 3.19.

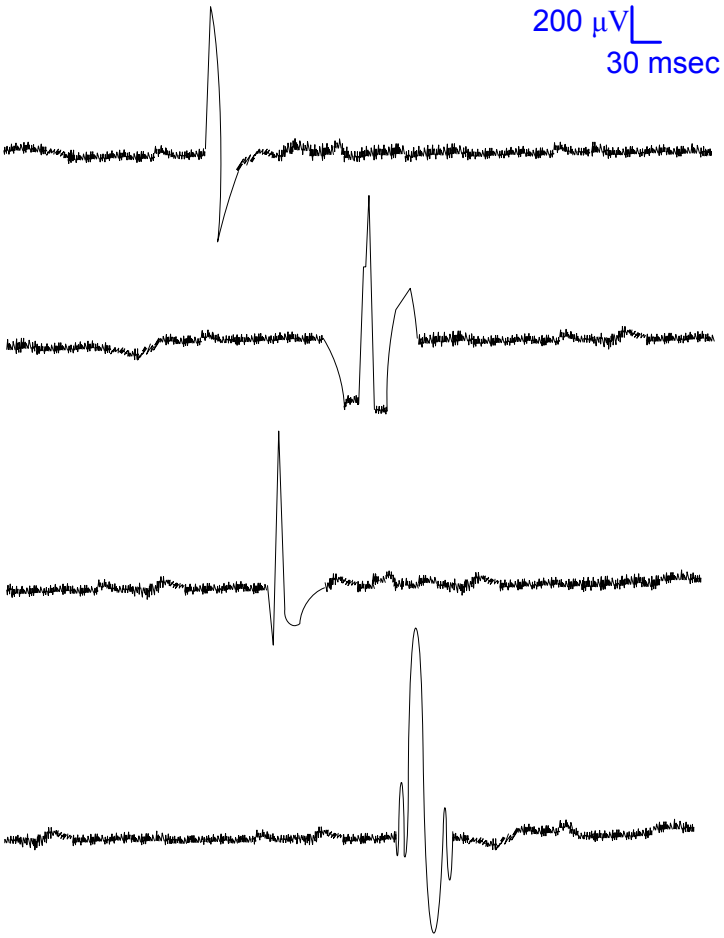

Figure 3.18 Fasciculations with different waveform shape and irregular frequency

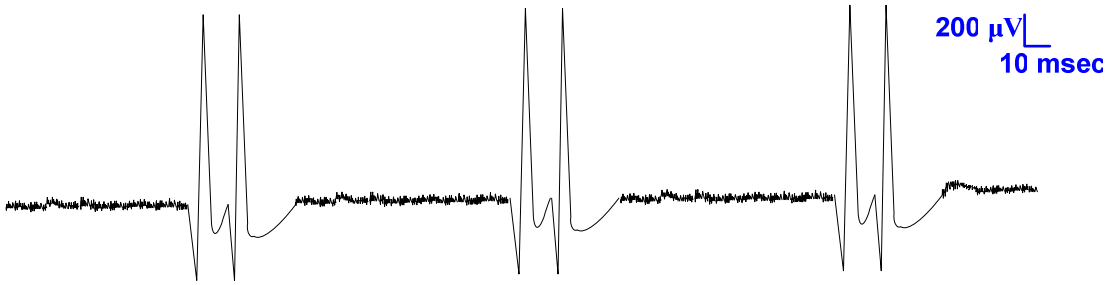

Figure 3.19 Doublets are the spontaneous discharges of motor units

3.4.2.3.2.3 Myokymic Discharges

These are the complex rhythmic, grouped, spontaneous repetitive discharges originating from the same motor unit [24, 26]. Myokymic discharges are characteristic with groups followed by a period of silence with subsequent repetition of a grouped discharge [10]. The firing frequency within the burst is 5 to 60 Hz [24]. The waveform of the myokymic discharges are shown in Figure 3.20.

Even though the pathophysiologic mechanism of myokymic discharges is unclear, there are numerous suggestion for the origin of these discharges such as abnormal excitability of lower motor neuron or peripheral nerves, spontaneous depolarization or ephaptic transmission of the demyelinated segments of nerve, the development of a rhythmic oscillating intraaxonal generator of action potentials [10, 24].

Clinically, they are characterized by involuntary flickering, rippling or undulating movement of a muscle [24, 36]. EMG is helpful in distinguishing myokymia from fasciculation, neuromyotonia and myoclonus [24, 36].

Myokymic discharges may occur in the limb is seen in the limbs in case of radiation-induced nerve damage especially in patients with a progressive plexopathy who have a prior history of cancer treated with radiotherapy. This situation is called radioation-induced plexopathy [10, 24, 26]. Furthermore, limb myokymia can be observed in radiation myelopathy, radiculopathy, entrapment neuropathy, spinal cord lesions associated with demyelination and gold intoxication, acute inflammatory polyradiculoneuropathy and multiple sclerosis [24]. Myokymic discharges in facial muscles occur in brainstem lesions associated with multiple sclerosis, neoplasms such as pontine gliomas and vascular diseases [10, 24]. Facial myokymia occurs 15% of the patients with Guillain-Barré syndrome, usually in the early stage of the illness [24]. In hypocalcemia induced by hyperventilation or with the use of acid-citrate-dextrose being an anticoagulant given during plasma exchange, myokymic bursts of the peripheral nerves can be provoked and can be decreased by the administration of calcium [24, 26].

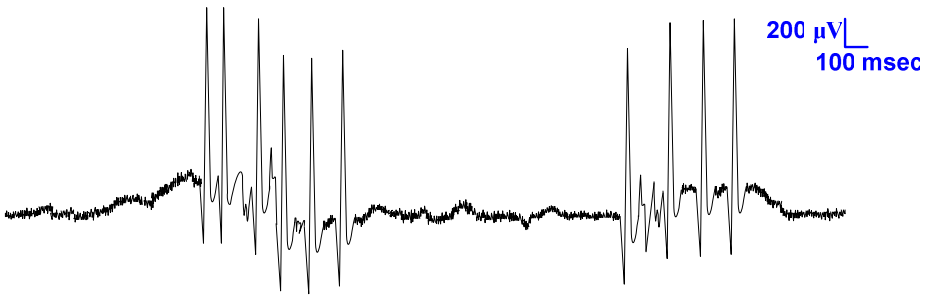

Figure 3.20 Myokymic discharges

3.4.2.3.2.4 Cramps

Cramps are involuntary, painful contractions of muscle when a muscle in shortened position and contracting [10, 24]. However, they are not a muscle phenomenon and originate from the discharges of motor axons in peripheral nerves [24, 26].

Cramp discharges are motor unit potentials firing repetitively and sometimes irregularly at high frequencies in the range of 200-300 Hz [24, 26]. The number of motor unit action potentials and their discharge frequency increase gradually and subsequently fall as the muscle contraction ceases [10]. Cramp discharges are represented in Figure 3.21.

Cramps may be benign such as nocturnal calf cramps and post-exercise cramps, or may be associated with many neuropathic, metabolic and endocrinologic conditions. They can resemble to contractures observed in metabolic disorders in clinical examinations. However, since the contracture is completely electrically silent the needle EMG appearance of the cramp discharges is different from that of the contracture [24].

3.4.2.3.2.5 Neuromyotonic Discharges

Neuromyotonic discharges result from the continous muscle fiber activity with peripheral origin, which is referred as neuromoytonia [26]. These discharges consist of burst of high-frequency, decrementing, repetitive discharges of a single motor unit with frequencies up to 300 Hz as shown in Figure 3.22 [10, 24].

Neuromyotonia is also referred as Isaac's syndrome, pseudomyotonia, neurotonia, normocalcemic tetany [24, 26]. Neuromyotonia arises from hyperexcitability of single peripheral motor axons either before or after they branch within the muscle [36]. Hence, the source generator varies from the proximal segment of the nerve to the intramuscular nerve terminals [26]. Neuromyotonia occurs in autoimmune anti-voltage gated potassium nerve axon channelopathy and Morvan's syndrome [24, 36].

The activity remains during sleep as well as spinal general or general anesthesia and is abolished by curare which can be considered as evidence for neural origin. Phenytoin and carbamazepine are also helpful in ceasing the activity [24].

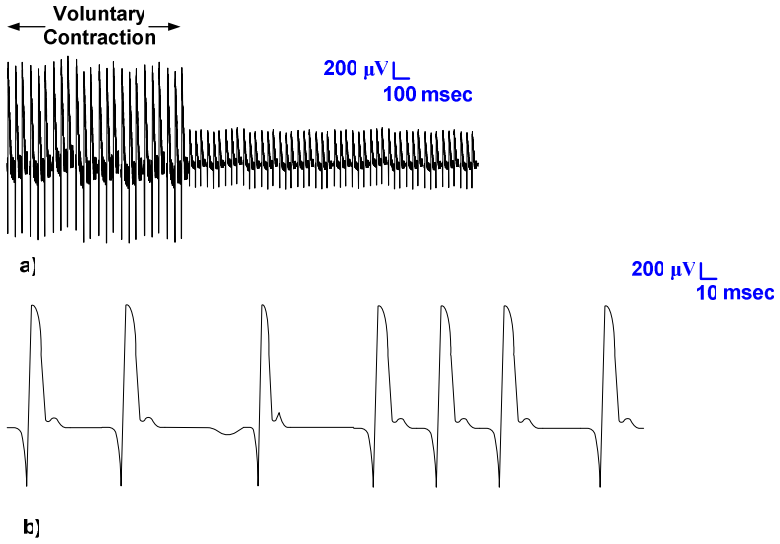

Figure 3.21 Cramp discharges. **a)** Cramp discharges seen in the relaxation phase **b)** The expanded view of the cramp discharges.

Neuromyotonic discharges may accompany to generalized stiffness, hyperhidrosis and delayed muscle relaxation after contraction. They may be seen in chronic neuropathic disease especially in old poliomyelitis and adult spinal muscular atrophy [24].

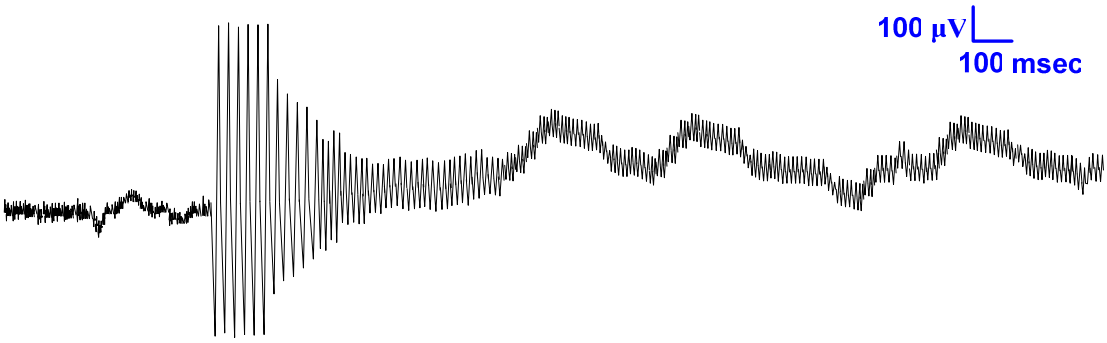

Figure 3.22 Neuromyotonic discharges from a single motor unit potential at high frequencies (150-250 Hz).

3.4.2.3.2.6 Rest Tremor

Tremor usually occurs during the voluntary contraction. However it can be also seen at rest as in case of Parkinson's disease and can complicate the interpretation of the spontaneous activity on EMG.

Tremor is recognized as a synchronous bursting pattern of MUAPs separated by relatively silent periods. Since it originates from simultaneous firing of multiple MUAPs the waveform is polyphasic as shown in Figure 3.23.

Rest tremor can be confused with the mykymic discharges. However, rest tremor can be differentiated from myokymic discharges through polyphasic shape of the rest tremor due to the contribution of many MUAPs. Nevertheless, myokymic wavform has a repetitive bursting pattern originating from the same motor unit. In addition, the amplitude of the tremor rises and falls where as that of the myokymic discharges remain relatively unchanged, when their pattern are examined closely [24]. The comparison between the rest tremor and the myokymic discharges in Figure 3.24.

3.4.3 Activities from Voluntary Contractions

These are the activities recorded during the voluntary contractions in varying levels. They can be classified as motor unit action potential (MUAP), recruitment pattern and interference pattern.

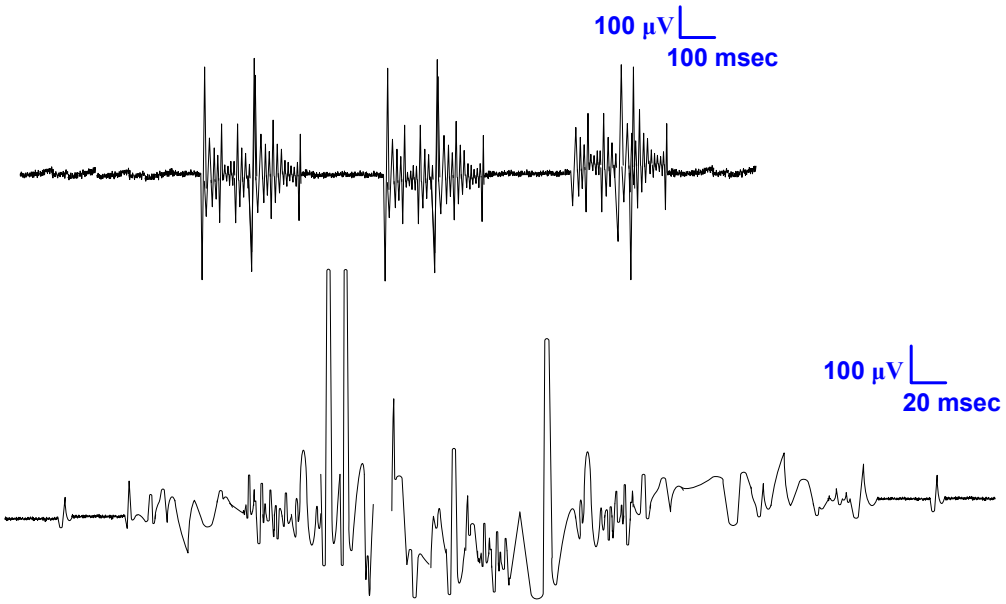

Figure 3.23 Rest Tremor as a bursting pattern separated by relatively silent periods

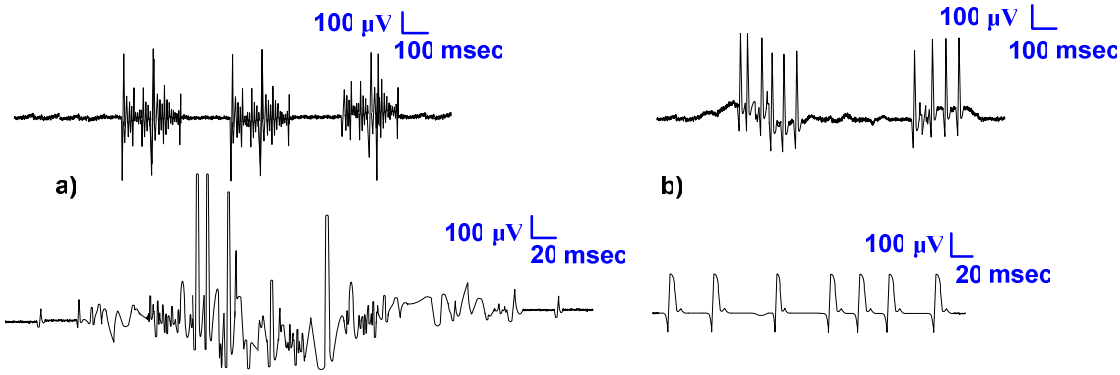

Figure 3.24 Tremor versus myokymia. **a)** Rest tremor is a polyphasic waveform due to the contribution of many MUAPs **b)** Myokymia yields from the repetitively firing of the same motor unit.

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3.4.3.1 Motor Unit Action Potential (MUAP)

As previously mentioned, skeletal muscle is organized functionally on the basis of the motor unit (MU). MU consists of the anterior horn cell, the motor neuron, the neuromuscular junction and the muscle fibers innervated by this motor neuron. It constitutes a bioelectric source located in a volume conductor consisting of other muscle fibers [8, 24, 50].

During the voluntary contraction, muscle fibers generate individually action potentials called single fiber action potentials (SFAPs) [7, 18, 27]. When a contraction is initiated or when an external stimulus is given, the motor unit generates an action potential which is the sum of electrical activities or that of SFAPs generated by all fibers belonging to any motor unit [24]. This action potential is called Motor Unit Action Potential (MUAP).

MUAP has quantitative parameters such as duration, amplitude, number of phase, number of turns, MUAP area, MUAP thickness, size index, satellite potential, spike duration, MUAP firing frequency (firing rate) and jiggle [51, 52].

The MUAP and its parameters are shown in Figure 3.25. Amplitude, duration, phases, and turns are previously mentioned. Other quantitative parameters will be discussed in the following paragraphs.

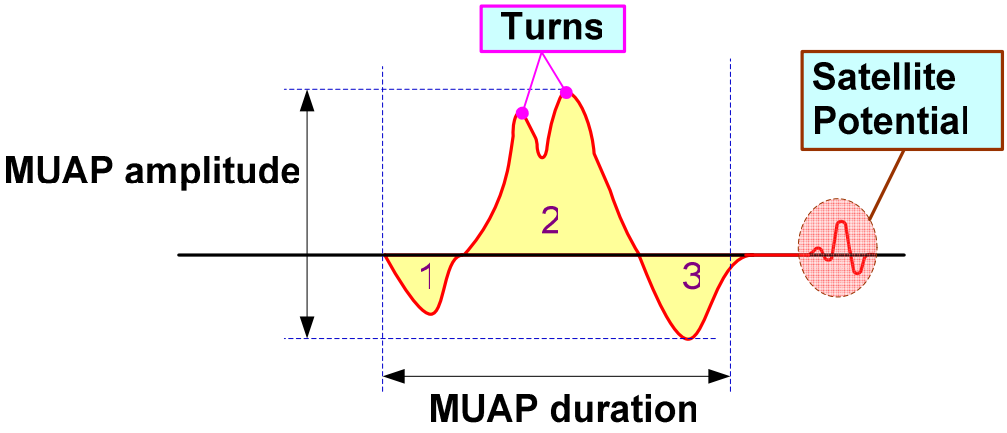


Figure 3.25 A triphasic MUAP with satellite potential

3.4.3.1.1 MUAP Area:

It is the area between the MUAP waveform and the baseline. It is related to both duration and amplitude [54]. Amplitude, duration and number of phases are three major parameters that characterize motor unit action potentials (MUAPs) in needle electromyography (EMG) [54]. The fact that the proximity of the electrode to the closest muscle fiber determines the amplitude has been shown by computer simulation and this makes difficult the differentiation of the neuropathic disorders from the myopathic disorders [26, 53, 54]. Thus, since greater number of muscle fibers lying within a 2 mm radius of the electrode tip contributes to this measure area measurements are less sensitive to measurement errors due to the needle placement and may help achieve this differentiation [26, 54].

3.4.3.1.2 MUAP Thickness:

It is the ratio between MUAP area and MUAP amplitude [26]. It is a parameter of MUAP which varies much less with changes in electrode potential [26]. This ratio which characterizes the 'thickness' of the potential is normalized by the amplitude in order to reduce to influence of the amplitude and is related to duration. MUAPs in myogenic cases are 'thin, the ratio of area/amplitude has usually lower values, even for atypical potentials with high amplitudes. MUAPs in neurogenic cases, even those with normal duration, have a larger area [54].

3.4.3.1.3 Size Index:

It is a relationship between MUAP thickness and MUAP amplitude. It is expressed as;

$$size\ index = 2.\log(MUAP\ amplitude) + \frac{MUAP\ area}{MUAP\ amplitude} \qquad (3.1)$$

This measure also evaluates the 'thickness' of the potential, but it has an advantage over MUAP thickness as a ratio between area and amplitude in that it is independent of the needle's movement. MUAPs are well distinguished from those of normal units by this index [53, 54].

3.4.3.1.4 Satellite Potential:

It is also known as late component, linked potential, coupled discharge or parasite potential. It is a small potential separated from the main MUAP by an isoelectric interval and firing in a time-locked relationship to main action potential [26]. This is a phenomenon seen in the early reinnervation. Following the denervation, muscle fibers are usually reinnervated by collateral sprouts from neighboring intact motor units. The newly constituted sprout is small, unmyelinated or thinly myelinated. Thus, their conduction velocity is slow. As a result of the slow conduction time and increased distance reinnervated muscle fibers generate time-locked satellite potential linked to the main MUAP. The sprout matures and the thickness of the myelin increases resulting in the rise in the conduction velocity. The satellite potential fires more closely and then becoming an additional phase or serration within the main MUAP [24]

3.4.3.1.5 MUAP Firing Frequency:

This is the number of firing of a MUAP in a second. The MUAP firing frequency (or firing rate) of the active units varies as the force increases beside the recruitment of the additional inactive motor units. Even though the firing frequency ranges from several Hz to 30 Hz during the early phases of the voluntary contraction, it seems to stabilize a wide range of forces [2, 24, 26]. The change in the firing frequency is referred as rate coding. The rate coding regulates the fine control at the beginning of the contraction [26].

3.4.3.1.6 Jiggle:

It is the alteration in MUAP configuration in each firing even though the needle electrode is not repositioned. It is referred as instable MUAP. It is an indicator for the

impairment in the function of the motor end-plate as seen in primary motor end-plate diseases or in immature motor end-plates [30].

3.4.3.2 Recruitment Pattern:

It is a potential observed in case of the contribution of the additional MUAP to the contraction and in the form of increase in MUAP frequency as the contraction intensity is increased [24]. When patients are requested to activate minimally a muscle a single or a few motor units are usually able to be recruited. Motor units have a recruitment frequency of 6–10 Hz [36].

3.4.3.3 Interference Pattern:

During the maximal contraction, many motor units begin to fire very rapidly. MUAPs normally overlap precluding recognition of individual motor unit potentials. This pattern which is recorded during maximal contraction is called interference pattern [10, 24, 26].

The spike density and the average amplitude of the summated response in interference pattern depend on a number of factors such as number of motor neurons capable of discharging, firing frequency of each motor unit, waveform of individual potentials [26].

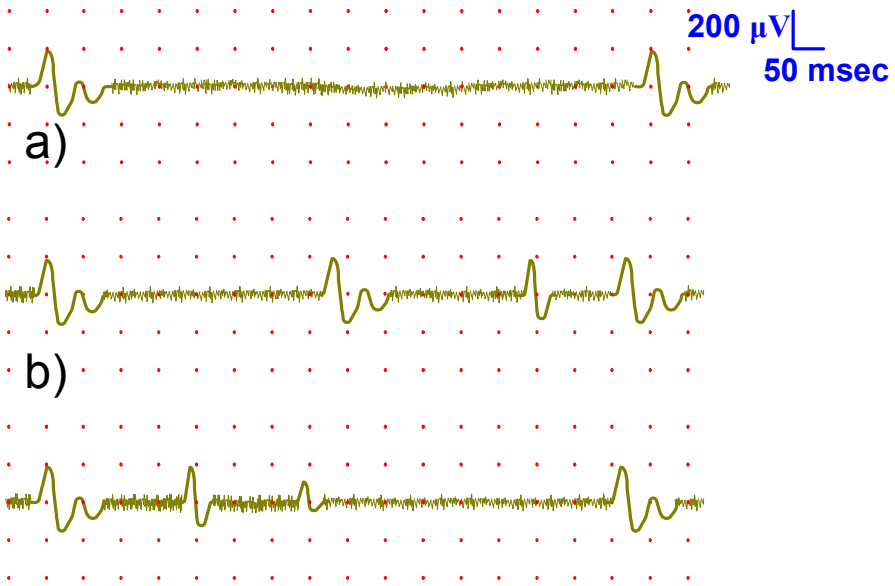


Figure 3.26 Normal Recruitment Pattern. **a)** With minimal effort of muscle contraction, a single motor unit is seen firing at 6 Hz. The time between 2 discharges is approximately 166 milliseconds (ms), corresponding to a firing rate of 6 Hz. **b)** Gradual increase in muscle strength results in recruitment of a second motor unit. Recruitment frequency 12 Hz, the recruitment interval is 85 ms. **c)**With further increase in muscle strength, a third motor unit is recruited.

In neurogenic cases, a reduced number of MUAPs fire at a higher frequency resulting in an incomplete interference pattern due to the reduced recruitment as shown in Figure 3.27-b. On the other hand, in myogenic cases, the number of MUAPs is normal however, the interference pattern consists of short-duration small-amplitude MUAPs as shown in Figure 3-27-c [24].

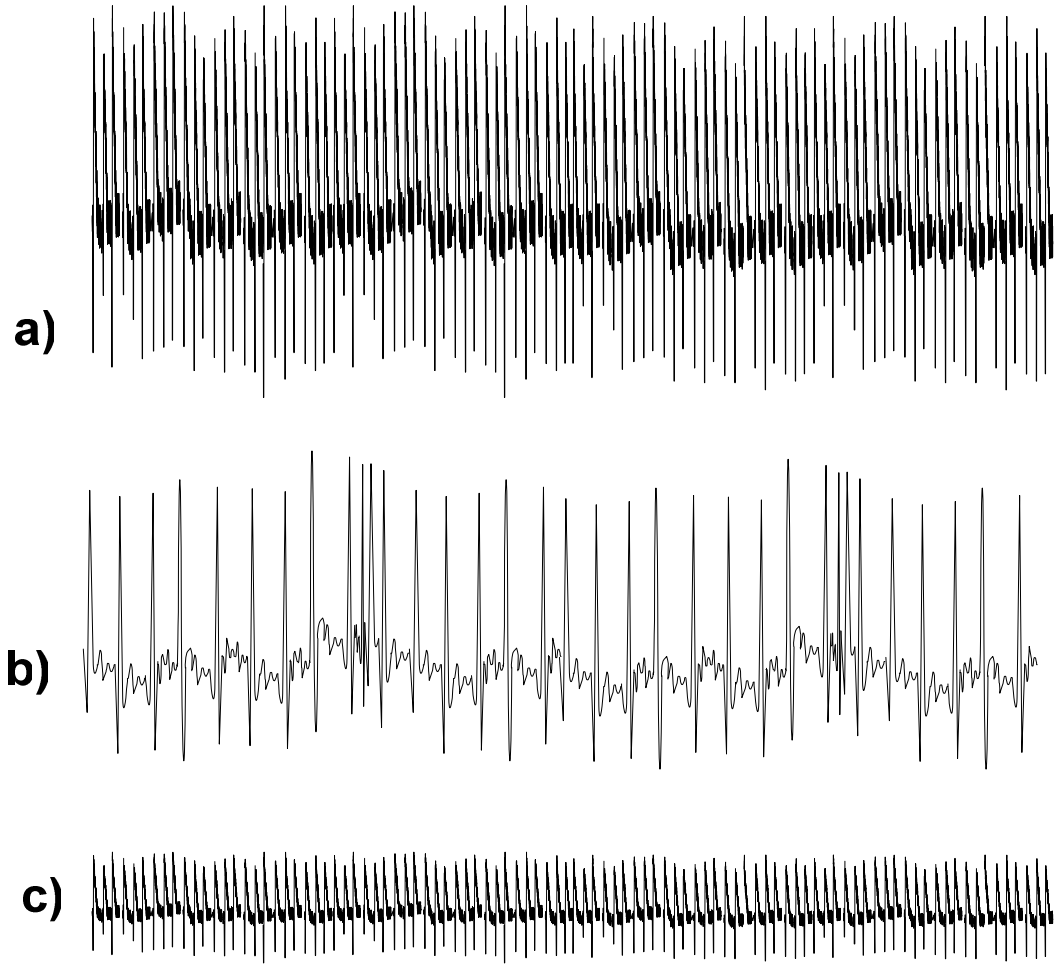


Figure 3.27 Interference patterns **a)** Normal: so many MUAPs fire during maximal contraction so that differentiating individual MUAPs is difficult **b)** Neurogenic: a reduced number of MUAPs fire at a higher frequency resulting in an incomplete interference pattern **c)** Myopathic: even though the number of MUAPs is normal, the interference pattern consists of short-duration small-amplitude MUAPs.

3.5 Electromyography (EMG) Techniques

3.5.1 Conventional EMG:

Conventional EMG is the most commonly electrophysiological technique that is routinely used in the clinical examinations of the neuromuscular disorders. This technique is used to record spontaneous, voluntary and occasionally stimulus-induced activity [38]. This is usually performed by using concentric needle electrode (CNE) or occasionally monopolar needle electrode (MNE) [30]. Electrodes of either type are inserted within a discharging motor unit territory [38].

The motor unit action potentials (MUAPs) are detected by this CNE. MUAPs are the sum of the single fiber action potentials (SFAPs) generated by the muscle fibers located within the uptake area of the CNE [30].

MUAP parameters such as amplitude, duration, number of phases, rise time that can be used in the differential diagnosis of various neuromuscular diseases are monitored by conventional EMG technique [3, 6, 26, 30].

3.5.2 Specific EMG Techniques

3.5.2.1 Single Fiber Electromyography (SFEMG)

When it is aimed to study physiological properties of individual muscle fibers, motor end-plates, terminal axon branches, a selective recording method is required [39]. SFEMG is a selective recording of action potentials from single muscle fibers with an uptake radius of about 300 μm during voluntary contraction [26, 30]. Single fiber EMG was developed to study the motor unit microphysiology in health and disease [30, 39].

3.5.2.1.1 Amplifier and Filter Settings:

The SFEMG electrode which is described in the preceding paragraphs ensures to record single muscle fiber action potentials selectively [26]. Due to its very small pickup range, the single-fiber electrode rarely records potentials from more than one or two fibers from the same motor unit [3]. Due to the small leading-off surface of the SFEMG electrode, bioelectric sources, which are located more than about 300 μm from the side port, will appear as common-mode signals and be suppressed by the differential amplifier [3, 26]. To further enhance the selectivity, the high-pass filter is set to 500 Hz to remove low-frequency volume-conducted background activity from distant fibers. The low-pass filter is set to 10 kHz in order to maintain the amplitude and the shape of the original spike [3, 26].

Due to the small recording surface of the active electrode, SFEMG electrode has a much higher electrical impedance than a concentric needle electrode (CNE) or monopolar needle electrode (MNE). Impedance values range on the order of several megaohns at 1

kHz for platinum needle [26]. Therefore, the amplifier must have very high input impedance in the order of 100 MΩ in order to maintain a higher signal-to-noise ratio [26, 39]. The common mode rejection ratio (CMRR) should be better than 200. The initial amplifier settings should be 0.2-1 mV/cm for sensitivity and 0.5-1 msec/cm for sweep speed [26].

3.5.2.1.2 Single-Fiber Potential:

The single-fiber potential recorded by a single-fiber electrode is a biphasic spike with a rise-time of 75 to 200 μsec. The total duration is about 1 msec [26]. Even though the peak-to-peak amplitude varies widely from 200 μV to 20 mV, it usually remains within the 1 to 7 mV [26, 38, 39]. The shape of the potential remains nearly constant with a time resolution of 5 to 10 μsec for successive discharges. The frequency spectrum lies within 100 Hz to 10 kHz with a peak at 1.61±0.30 kHz [26].

3.5.2.1.3 Recording Procedures:

Potentials for SFEMG can be generated by either electrical stimulation or voluntary activation.

In voluntary activation, the electrode is placed in such a position that action potentials from two muscle fibers belonging to the same motor unit are recorded. The interpotential interval (IPI) between the individual action potentials obtained from the two muscle fibers varies from discharge to discharge. This variability is referred as to jitter [30]. This procedure is called voluntary single fiber electromyography (v-SFEMG) [55].

For electrical stimulation, intramuscular nerve twigs or small extra-muscular nerve branches may be stimulated with a monopolar needle electrode. Stimulus strength may be adjusted to stimulate very few axons. Thus recording can be made from just one of a few muscle fibers. The jitter measured between the stimulus and the action potential of the responding fiber reflects neuromuscular transmission. Therefore, only a single motor end-plate is studied [30]. This procedure is referred to as stimulation single-fiber electromyography (s-SFEMG) [56]. It is different from the method in voluntary activated muscle requiring recording from a pair of muscle fibers. This method allows the detailed study of neuromuscular transmission during different firing rates and does not require the co-operation of the patients. The technique can therefore be used especially in children, unconscious patients and other patients that are unable to cooperate [30].

3.5.2.1.4 SFEMG Parameters:

SFEMG is used to study physiological and morphological parameters of motor unit and its constituents [38].

Fiber Density (FD):

The distribution of muscle fibers within the muscle can be quantified with SFEMG recordings [30]. FD describes the degree of grouping of muscle fibers of a single motor unit in the cross-section of a muscle [38]. FD can be defined as the mean number of associated single-fiber potentials that fire almost synchronously with the initially identified potential [26, 38].

An electrode is inserted into a slightly activated muscle and a position is sought in which the action potential from one muscle fiber is recorded with maximal amplitude [30]. This potential is used to trigger the sweep and the number of synchronous action potentials with a rise time less than 300 μs and amplitude of greater than >200μV are counted in 20 different electrode positions. The high-pass filter is set to 500 Hz. Fiber density is calculated by the average of these 20 values [26, 30, 39].

It is a measure of the number of muscle fibers from one motor unit within the electrode uptake radius of about 300μm. In the normal muscle FD is around 1.5, depending on the muscle. In old age the FD in distal muscles increases [30]. By definition, the possible lowest value is 1 [26].

FD is increased after collateral sprouting in neuropathies due to the reinnervation [30, 39]. Three to ten muscle fibers from the same motor unit can be recorded in case of reinnervation [39] However, FD can also be increased in many myopathies which indicates small grouping of fibers, e.g. due to fiber splitting or regeneration. This parameter is very sensitive to detect pathology in the organization of the MU but additional studies are necessary for exact definition of the type of pathology [30].

FD is typically used for the detection of slight changes in the motor unit topography for example in reinnervation or in dystrophies [30].

Jitter and Impulse Blocking:

Neuromuscular transmission yields from the sequential events occurring in the neuromuscular junction initiated by the arrival of the nerve action potential such as influx

of calcium from opening channels at the nerve terminal, release of quanta of acetylcholine (ACh) from the presynatic membrane, binding of ACh to ACh receptors on the post-synaptic membrane, structural transformation of sodium channels, depolarization of the sarcolemma, generation of EPPs and triggering of AP [39]. There is latency between the stimulation of nerve AP and the propagation of muscle fiber AP. The variability in this latency is called jitter [26]. Jitter which is also referred to as interpotential interval (IPI) reflects the variability of the neuromuscular transmission time in the two motor endplates, and is a measure of the safety factor in the motor end-plate, which is the ratio between the EPP and depolarization threshold [38, 30]. It is normally 5 to 50 μsec [3, 30]. Jitter can be studied either during voluntary activation or with electrical stimulation [30, 57].

In voluntary activation, the electrode is placed in such a position that APs from two muscle fibers belonging to the same motor unit are recorded [58]. The interpotential interval (IPI) between the individual action potentials obtained from the two muscle fibers varies from discharge to discharge [30]. In electrical stimulation, intramuscular nerve twigs (in most muscles) or small extra-muscular nerve branches (n. facialis branches to m. orbicularis oculi) may be stimulated with a monopolar needle electrode. Stimulus strength may be adjusted to stimulate very few axons and recording can be made from just one of a few muscle fibers. Neuromuscular transmission is reflected in the jitter, which is measured between the stimulus and the action potential of the responding fiber, hence only a single motor end-plate is studied. This is different from the method in voluntary activated muscle, which requires recording from a pair of muscle fibers. This method thus allows the detailed study of neuromuscular transmission during different firing rates and does not require the co-operation of the patients [59]. The technique can therefore be used especially in children, unconscious patients and other patients that are unable to cooperate [30, 39, 60, 61].

Jitter analysis is a useful and sensitive parameter in the diagnosis of patients with neuromuscular transmission disorders. Jitter is not only increased in myasthenia gravis, but also in ongoing reinnervation, botulinum intoxication and during regeneration in myopathies [62, 63].

3.5.2.1.5 SFEMG Indications:

SFEMG is used as a complementary technique to the conventional electromyographic investigation in selected patients. It is the most sensitive technique to study abnormalities of neuromuscular transmission [65]. FD is typically used for the detection of slight changes in the motor unit topography as in reinnervation as in case of anterior horn cell disorders or in dystrophies such as Duchenne and Becker dystrophy, limb-girdle dystrophy. Jitter is used to reveal disturbed neuromuscular transmission as in case of myasthenia gravis, Lambert-Eaton Myasthenic Syndrome and Botulism [30, 39, 63, 64].

Furthermore, SFEMG is accepted as the most sensitive method in the diagnosis of the myasthenia gravis [66-68]. It is considered as an important tool in the assessment of the neuromuscular diseases [69].

There is a study about that stimulation single-fiber EMG (s-SFEMG) might be beneficial in the early detection of the NMJ transmission blocking in myasthenia syndrome following acute insecticide poisoning [59, 70].

3.5.2.2 Macro Electromyography (Macro EMG)

Concentric needle electrode with an up-take area of 500-μm radius and monopolar needle electrode covering an area with a radius of about 1 mm are able to record electrical activity from wider zone compared with SFEMG electrode with a 300-μm radius. However, CNE and MNE are still giving information only about a part of a motor unit territory [26].

To capture the total electrical activity generated by a motor unit, a technique using an electrode with a much greater recording surface is required. This technique is called macro electromyography (macro EMG) [26]. This technique is schematized in Figure 3.25.

The purpose of the macro EMG technique is to record the macro MUAP being defined as the electrical activity of a majority of muscle fibers of the entire motor unit [38, 71]. Hence, it can be considered as a technique that may provide information about the size of motor unit by reflecting the summated activity of the entire MU [8, 10]. The Macro EMG recording is non-selective and most fibers from the entire motor units contribute to the signal being a compound action potential. The peak-to-peak amplitude and area of the Macro EMG signal is correlated with the number and size of muscle fibers in the motor unit [30-38].

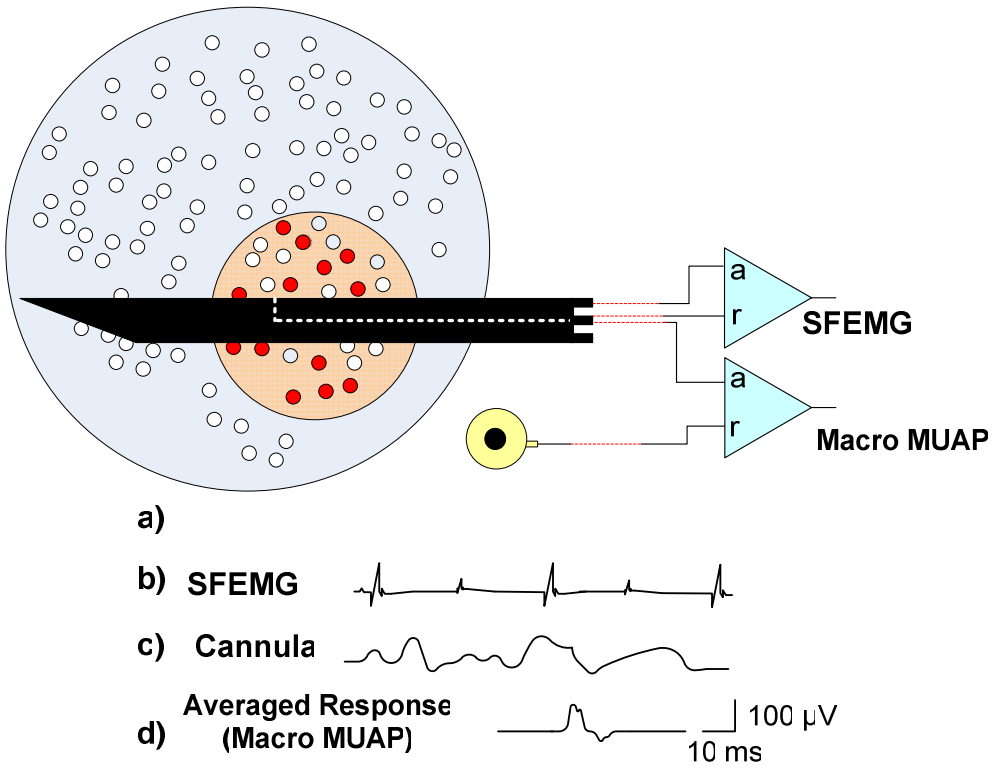


Figure 3.28 a) Macro EMG recording used to trigger the traces and the cannula referenced to a distant electrode records activity of most muscle fibers within the motor unit (marked with red) **b)** SFEMG signal taken by the Single Fiber electrode inserted inside the cannula **c)** The potential of the cannula where non-synchronized activity is also included **d)** Averaged Macro MUAP (non-synchronized is eliminated) (a: active surface r: reference)

Macro EMG is based on the use of Macro needle electrode. Recordings are made between a large segment of the cannula of this needle and a distant surface reference electrode [26, 30,38]. Macro electrode uses a modified SFEMG electrode to trigger on an action potential from a nearby muscle fiber [10, 26, 38]. This action potential is drawn to one amplifier channel. On the other, the time-locked macro MUAP recorded by the cannula of the electrode is drawn to a second channel. The activity of the neighboring motor units which are not time-locked to the SFEMG signal are eliminated as background noise during signal averaging [10, 26, 38, 72].

The electrode is inserted into the voluntarily activated muscle and a position is sought where an acceptable SFEMG potential is seen. The activity from the cannula is averaged until a smooth baseline and a constant Macro MUP is obtained [30, 72].

3.5.2.3 Scanning Electromyography:

Scanning EMG is an experimental technique developed to study the spatial and temporal properties of a motor unit electrical activity in order to better understand the topography of a motor unit [10, 30, 39].

It has been used in several studies alone or in combination with oher techniques to identify the electrophysiological events in the motor unit that generate the individual parts of the MUP, to make possible a description of the microphysiology and anatomical arrangement of muscle fibers in normal and abnormal MUs, as well as the motor unit size [73-75].

Scanning EMG was first performed by Stålberg and Antoni in 1980 in order to investigate the MU organization. An electrophysiological cross-section of the MU was obtained for this purpose [17].

Hilton-Brown and Stålberg used Scanning EMG method in conjunction with SFEMG in patients with muscular dystrophies (MDs). The aim of this study was to reveal that the EMG changes yield from not only the loss of muscle fiber but also a neurogenic component due to the regenerative process. By means of the SFEMG, they investigated the alterations in parameters such as fiber density (FD) and jitter. In scanning EMG, they

demonstrated local fiber grouping resulting in silent areas within the motor unit territories supporting the idea of rearrangement of muscle fibres within the motor unit [76].

In another study, Hilton-Brown and Stålberg used Scanning EMG method in conjunction with Macro-EMG in patients with MDs to investigate the distribution of activity within the Mu territory. They studied fiber density and the total size of motor units in muscular dystrophy. In contrary to CNEMG findings that suggest smaller MUs in this myopathic disorder, they found normal or slightly reduced Macro-MUAP in Macro-EMG and normal length of cross-section in Scanning EMG. In addition, Scanning EMG revealed local fiber grouping mixed with silent areas demonstrating that the findings of Macro-EMG were due to the degeneration and subsequent regeneration [77].

Stålberg and Eriksson was studied mandibular motor system previously by using Scanning EMG method. They demonstrated that mean length of cross-section of the masseter muscle was less than that of large muscles. This confirmed that MUs of the muscles performing fine adjustment of jaw movement such as masseter have smaller MU territories and they contain fewer muscle fibers. On the other hand they found that muscles important in force development of the mandibular motor system as in biting in intercuspal position or opposing gravity have larger MU territories [78].

In a previous study, Stålberg and Diószeghy performed recordings from the tibialis anterior and biceps brachialis muscles of the healthy, neurogenic and myopathic subjects and compared parameters that can be measured by scanning EMG between these three groups in order to verify the rearrangement of muscle fibers in abnormal muscles [17].

Gootzen, Vingerhoets and Stegeman used the scanning EMG method to investigate the structure of human quadriceps musle motor units. They conducted this study with one

group of healthy subjects and two groups of patients with either neurogenic or myopathic disorders. Two parameters were defined. One of them was *S* defining a spatial measure of the motor unit territory. The other parameter was *T* being the temporal measure of the time dispersion between MUAPs. In conclusion, territories of the myopathic patients were smaller where as those of neurogenic patients were larger. Furthermore, a parameter measuring the "integrity" of the motor unit was proposed to describe the variation in action potential propagation velocity [79]

In this technique, two distinct electrodes are used. One of them is used to record single fiber action potential from a muscle fiber belonging to the motor unit under the investigation as a triggering signal [30, 39]. This electrode is usually a SFEMG electrode, but since it is much cheaper, it is disposable and it requires a lower contraction level, a CNE can be also used by adjusting the high-pass filter to 2 kHz instead of a SFEMG electrode [66]. This SFAP is used to select time-locked electrical activities with this potential which also belong to the motor unit under investigation during the recording process. It is inserted into the muscle during a slight muscle contraction and is positioned so that a low-threshold potential is recorded [10]. The second electrode is used to record MUAPs from different locations of the motor unit in question during this recording process. This is a CNE and is inserted into the same motor unit as the SFEMG electrode. The correct position for this electrode is found by observing the synchronous activity recorded by CNE with that of SFEMG on the oscilloscope or on the display of the EMG instrument [10, 30, 38]. The scanning electrode is then pushed trough the MU after which the scanning is started by pulling the scanning electrode in 50-μm steps or multiples thereof trough the MU by a linear actuator [10, 16, 17, 30, 38]. In this way an electrophysiological cross section of the MU is obtained [17, 30].

In addition to the parameters used with conventional EMG such as amplitude and duration, new parameters are introduced with Scanning EMG. These are length of MU

cross-section, fractions of MUs, silent periods, polyphasic and complex portions of MUAPs, maximum duration and the maximum amplitude [16, 17].

The length of MU cross-section is defined as the distance between the first and the last signals along the recording corridor.

The fractions of MUs produce the spiky part of the MUAP. They are separated by low-voltage sections (<50μV). They reflect the activity from groups of muscle fibers innervated by the same axon branch and have end-plates in a limited region.

The silent periods are the sections in electrical activity with peak-to-peak amplitude less than 50 μV. The silent periods separate the fractions of MU activity along the path of the upward movement of the scanning (recording) electrode. They can be due to the proliferation of the fat or connective tissue in the muscle during the degenerative process.

The polyphasic and complex portions of MUAP are detected by conventional EMG only by chance. In Scanning EMG they are revealed anywhere along the path of upward movement of the scanning electrode. The maximum duration is the real duration of MUAP. The maximum amplitude is the real maximum peak-to-peak amplitude of the MUAP in a motor unit territory [8, 16, 17].

In moderate or pronounced neuropathies the territory of the MU is essentially unchanged but the FD within the MU is increased. Most of collateral sprouting takes place only within the territory where the MU was originally represented and does not extend to new fascicles [8, 16, 17, 30].

In myopathy, the increased fragmentation of motor units, the larger number of fractions, the presence of silent areas, and the increase in the number and length of polyphasic segments support the concept of motor unit reorganization, which is caused by the proliferation of fat and connective tissue as well as degenerative and regenerative processes [8]. Silent areas are commonly seen due to loss of muscle fibers [30]. Sections with polyphasic MUAPs are common in myopathic motor units [30].

Scanning EMG has not been used in routine clinical examinations but has served as a research tool for better understanding of the motor unit morphology and the generation of EMG signals in different parts of the motor unit in particular [30].

3.5.2.4 Quantitative EMG Techniques:

3.5.2.4.1 Interference Pattern Analysis (IPA)

An increase in force from zero up to maximum voluntary contraction (MVC) leads the increase in number of spikes and amplitude in the EMG. Individual MUAPs can be identified at weak effort, whereas at higher efforts, there is interference with summation and cancellation of these action potential components [80, 81].

As previously mentioned, interference pattern is determined by several factors including descending input from the cerebral cortex, number of motor neurons capable of discharging, number of recruited motor unit, firing frequency of each contributing motor unit. Despite the complexity resulting from such factors, interference pattern analysis provides a quantitative measure to evaluate the relationship between the number of firing

motor unit and the muscle force exerted with 10-100% of the maximal voluntary contraction [26, 82, 83].

Interference of MUAPs is based on their summation, an overlap already starting at 10% MVC at some sites of the muscle and is present at all sites at 30% MVC [82]. The number of spikes per unit time increases with the increasing force up to 2 kg in biceps brachialis muscle. These spikes being influenced by the number of MUs recruited and their firing rate up to 50% of the MVC increase up to 50% of this contraction level and then, remain constant due to interference of APs with cancellation of small spikes [80].

IPA is useful in assessing the orderly recruitment of MUs. The recruitment is assessed by observing the "fullness" of the IP, which reflects the number and firing rates of the component MUs, and the amplitude of the IP signals [81].

Interference pattern analysis (IPA) of the electromyogram (EMG) is a useful tool in describing the muscle activity, the muscle fatigue, EMG changes in occupational work, in the diagnosis and classification of chronic muscle pain, in monitoring the disused muscle, and dystonic muscles treated with botulinum toxin and in the diagnosis of patients with neuromuscular disorders [80].

Motor unit potential analysis may give information only for pathophysiological and physiological changes in motor units recruited at weak effort, whereas IPA may reflect changes in motor units recruited in the whole force range [80].

There exist several methods developed for interference pattern analysis such as decomposition, power spectrum analysis (PSA), turn/amplitude analysis (TAA), expert quantitative interference pattern (EQUIP) analysis [80, 82].

3.5.2.4.1.1 Decomposition:

It is an automated analysis method that requires special software. Decomposition techniques break down the IP into its constituents, the individual MUAPs. Using a four-surface needle multi-electrode, the precision decomposition technique extracts a MUAP by simultaneous recording of the signal from different pairings of the four recording surfaces. Under such conditions, the MUAP gets a unique signature that accurately distinguishes it from other MUAPs. Evaluated variables are the MU firing frequency, MUAP duration, MUAP amplitude, MUAP area and percent polyphasia [52, 82].

3.5.2.4.1.2 Power Spectral Analysis (PSA):

This is a technique where IP is decomposed into harmonically related sinusoidal-waves of different phases, frequency and amplitude, either by analogue octave-band filtering (early PSA) or fast Fourier transformation using anti-aliasing filters and a Hanning window [82]. Then, the power spectrum of these harmonics is computed. Median and mean frequency values can be calculated for the IP in question [80].

The highest peak seen during the maximal contraction lies within a frequency range of 100 to 200 Hz in normal subjects [26]. The peak shifts to higher frequencies in patients with myopathy and to lower frequencies in patients with anterior horn cell lesions [26, 80, 84].

3.5.2.4.1.3 Turns-amplitude analysis (TAA):

Turns-amplitude analysis is a quantitative technique used to evaluate the interference pattern (IP). It is achieved by measuring the number of turns per second (T/S) of an EMG signal and relating this parameter to the mean change in amplitude per turn (MA or A/T) [85]. A turn is defined as the directional change of the EMG signal when the amplitude difference is greater than 100 μV between these changes in polarity [26] [81, 83, 85-88]. When the force of contraction increases, more and larger motor units are recruited and the potentials of the signals generated by these motor units are summated. Therefore, mean amplitude (MA) of the interference pattern increases with force of contraction to a maximum value. However, the number of turns per second (T/S) initially rises and then remains the same at higher force levels [85].

The parameters evaluated by TAA are turns per second (T/S), amplitude per turn (A/T), T/S:A/T, duration between turns, upper centile amplitude (UCA), number of small segment (NSS), activity [82-84]. T/S is the number of change in polarity of the EMG signal provided that the deflection exceeds certain level such as 100 μV [10, 26, 85]. A/T is the mean change in amplitude (i.e. mean amplitude MA) per turn [85]. As implied by its name, duration between turns is the time interval between turns which gives the distribution of time intervals between spikes [10]. UCA which is also referred as envelope amplitude is defined as the amplitude difference between two turns, above which only 1% of higher amplitude differences can be found. It quantifies the amplitude of the largest spikes of the interference pattern [84]. NSS is number of small time intervals between turns excluding turns obtained between MUAPs [84]. It is defined as defined as the number of segments less than 3 ms in duration and 0.5–2 mV in amplitude and that of segments less than 1.5 ms in duration and less than 0.5 mV in amplitude [82]. The activity expresses the time within an epoch where MUAPs are present. Activity should satisfy the following criteria: duration less than 1.5 ms and amplitude less than 0.5

mV, duration less than 3 ms and amplitude of 0.5–2 mV, and duration less than 5 ms and amplitude less than 2 mV [82].

Basically, TAA is an IPA method for the quantitative analysis of the pattern of electrical activity of muscle during contraction against a standard load which is usually of 2 kg in order to distinguish patients with myopathy and normal subjects [10]. As the force of contraction increases in normal muscles, the number of turns per second (T/S) and the mean amplitude (MA) also increase. It has been reported that the T/S in the normal IP measured in biceps muscle becomes relatively constant or slightly decreases when the force of contraction is greater than 30%-50% of maximum, whereas the MA continues to rise at these force levels [89]. This analysis has been indicated increased number of turns/second in most patients with myopathy, in some patients associated with decreased mean amplitude per turn [84]. The idea behind this analysis method is that number of T/S is influenced by the number of motor units and by their firing frequency. Thus, summation due to the overlap of MUAPs influences both number of turns and amplitude. As the summation increases (i.e. increase of force of contraction), more cancellation will occur resulting in the higher mean amplitude of the interference pattern [81, 84].

TAA at a fixed relative maximum force such as 30% of maximum voluntary contraction (MVC) in patients with either myopathic or neurogenic disorder demonstrated that this method has been better in differentiating myopathic, neurogenic and normal muscle than analysis at a force of 2 kg [84, 91]. Nevertheless, the maximum force of voluntary contraction exhibits variations between individuals and disease states [85]. Besides, TAA at a given force level requires cooperation of the patient. In addition, determination of the adequate force level is time-consuming [84]. Furthermore, since they do not act on a lever or tendon, the force measurements are difficult for the muscles of facial expression [85]. Due to the limited utility of the TAA with force measurement, several modifications in this method have been suggested in order to eliminate the estimation of muscle force [10, 84].

To perform the TAA without force monitoring, the cloud methods such as T/S–A/T cloud method, activity-UCA-NSS cloud method have been developed. Normal Cloud is an area that contains more than 90% of the data points obtained from studies made in normal muscle [90]. Therefore, a plot of mean amplitude as a function of turns can be obtained [82, 84]. In order to form T/S-A/T cloud, interference patterns are recorded from different 6-10 sites at various 3-5 different force levels which range from zero to maximum. Cloud of normal values is constructed by computing the log(A/T-100) and log(T/S). Two lines at the mean log (A/T-100) ± 2 SD being parallel to the regression line between log(A/T-100) and log(T/S) provide the curvilinear upper and lower limit of the cloud. These limits are constructed again when plotting T/S and A/T values on linear scales. T/S value which exceeds 1% of the T/S values is taken as the upper reference limit. A virtual horizontal line drawn through the highest A/T value is used to complete the cloud. The normal cloud includes 90% of the normal values. An individual result is considered as abnormal if more than 10% of the values fall outside the normal boundaries [82].

As shown in Figure 3.26-a, the normal values are distributed inside the cloud where the boundaries are built as described as above. In neurogenic cases, collateral sprouts due reinnervation increase the amplitude. Besides, the number of turns decreases due to the motor unit loss. Thus, the distribution of values is located in favor of the amplitude as represented in Figure 3.26-b. In myopathic cases, mean amplitude is decreased. On the other hand, since number of motor units does not change and also recruitment pattern dominates, number of turns is increased. In addition, multi-serrated polyphasic MUAPs contribute to the increase in number of turns. Therefore, a shift of the distribution of values occurs outward the cloud in favor of turns as illustrated in Figure 3.26-c [80, 88, 90].

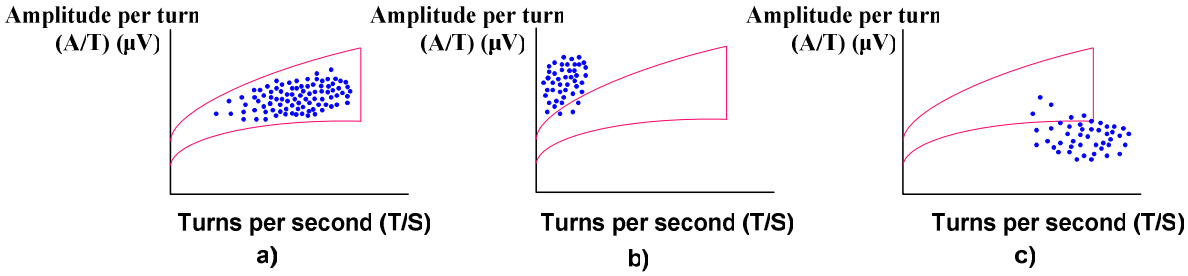


Figure 3.29 Cloud in turn-amplitude analysis: **a)** normal values **b)** neurogenic case **c)** myopatic case

The shape of the normal turns-amplitude cloud is affected by the maximum effort at which recordings are made. In strong normal subjects those clouds may falsely indicate a neurogenic abnormality when the subjects exert near maximum force [90].

3.5.2.4.1.4 Peak Ratio Analysis (PRA):

Peak ratio can be defined as the maximum value of the ratio of turns per second (T/S) to the mean amplitude (MA). It is obtained by using the mean amplitude per turn (A/T). Since it increases linearly with force, it is used as an indicator of force [80, 92]. Peak ratio analysis is a TAA performed without the requirement for force monitoring [93]

The electrical activity is recorded during gradually increasing contraction from zero to maximum voluntary contraction (MVC) in steps of 100 milliseconds over 10-20 seconds. The ratio T/S:A/T is calculated for each segment. The ratio values are then sorted regarding the increasing corresponding A/T values. A mean value of 10 consecutive, sorted ratio values (equal to 1 s) is calculated in order to smooth the data points of high variability. This procedure is repeated 100 times in steps of 100 ms to gain a moving average. From the 1-second segment of the moving average containing the

highest T/S:A/T value which is the peak-ratio, the variables T/S, A/T, and time intervals between turns with a duration of 0–1.5 ms, 1.5–5 ms and 5–20 ms are calculated. For the number of time intervals, a logarithmic transformation is further performed in order to obtain a normal distribution [82, 84].

The peak ratio is measured in at least 10 sites in each muscle in order to obtain representative values. As the interference pattern varies in different parts of the muscle, these 10 sites should be carefully distributed in the muscle using at least 3 insertions in each muscle. At each site in the muscle, the peak ratio is obtained from the interference pattern during a gradual increase in force from zero with an on-line display of a plot of ratio of turns to mean amplitude against mean amplitude [84].

At a moderate force of about 10-30% of the maximum voluntary contraction the ratio will reach a maximum value but the mean amplitude will continue to rise. The peak ratio is obtained at the moment when the ratio begins to decline where as mean amplitude continues to increase during the gradual increase in contraction [84].

The sensitivity of the method is related to the structural changes of the motor units in these disorders. In neuropathy patients with abundant spontaneous activity, peak-ratio parameters may change to normal, thus leading to false negative results [92].

3.5.2.4.1.5 EQUIP (Expert Quantitative Interference Pattern) Analysis:

EQUIP analysis is also referred as activity/UCA/NSS cloud method, interference patterns are recorded by the same procedure as that used for the T/S–A/T cloud method. In the new version of the EQUIP analysis, UCA is replaced by the amplitude of the IP

envelope that reflects the amplitude of the largest MUAPs in spite of the summation of small MUAPs [80, 81]. Upper centile amplitude (UCA), measures the largest amplitude change between successive turns after excluding the 1% largest values within the analyzed epoch [81, 94]. The cloud in the method is constructed in a way similar to the T/S–A/T cloud. The difference is that 3 standard deviation (SD) is used instead of 2 SD in determining the curvilinear upper and lower limit of the cloud [82]. Thus, three parameters are measured by EQUIP analysis [88];

Activity: Activity corresponds to the time within an epoch during which motor unit action potentials are present. In other word, activity quantifies the fullness of the IP [97]. Activity is the sum of the duration of segments or time intervals between turns below the duration of 1.5 to 3.0 ms, depending on the amplitude [80]. It indicates the duration of the EMG activity which is present in a 1-second epoch. If the activity is equal or greater than 500 milliseconds, it is considered as complete interference pattern otherwise they are considered as incomplete interference pattern [81, 94].

Amplitude of the IP envelope (AIPE): It can be defined as the amplitude between lines joining either the negative peaks or the positive peaks. It increases in neuropathies where as it decreases in myopathies. The envelope amplitude quantifies the amplitude of the largest spikes of the IP [80].

Number of Small Segments (NSS): A segment or time interval is the portion of the signal between two successive turns [94-96]. NSS is the number of small-amplitude fluctuations in baseline that cannot be considered as turns. NSS corresponds to number of turns obtained from voltage variation within the motor unit potentials, excluding turns obtained between motor unit potentials. The number of small segments is the sum of the segments below a certain duration, depending on the amplitude, e.g., less than 1.5 ms for

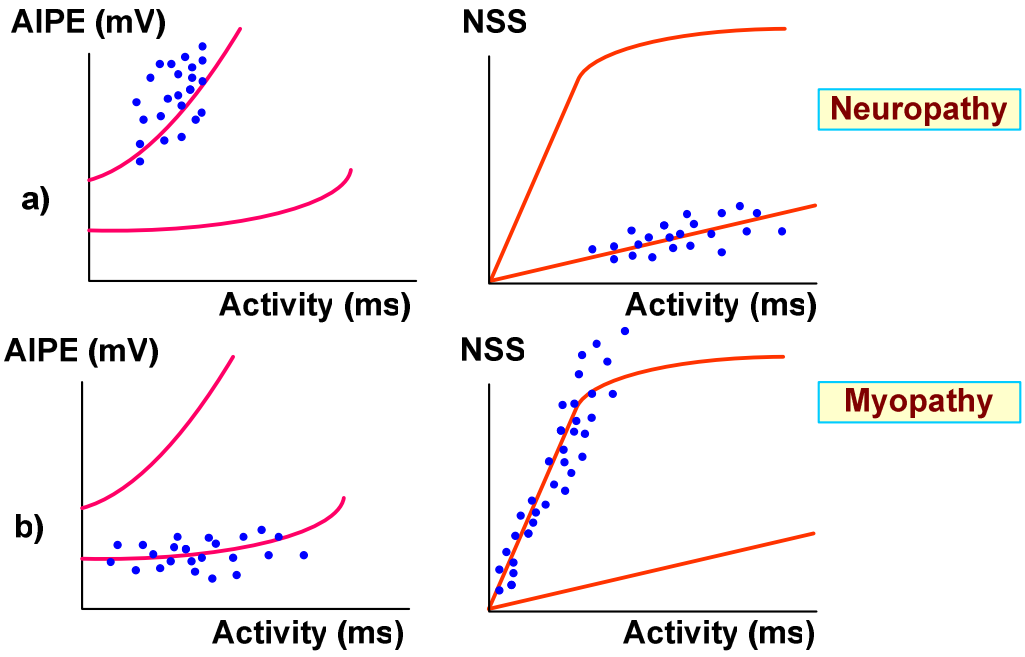


Figure 3.30 The results of EQUIP analysis in neuropathy and myopathy (AIPE=Amplitude of the interference pattern envelope; NSS=Number of small segments)

segments with amplitude of up to 0.5 mV [80]. Pratically, it is the number of segments with amplitude less than 2 niV that are included in the activity measurement [94]. It measures the complexity of the IP, which includes the polyphasicity of the MUAPs, among other factors [97].

Since the activity/APEI and activity/NSS clouds each represent 90% of the normal data points, an individual result is regarded abnormal if more than 10% of the individual values fall outside one or both of them [82].

As demonstrated in Figure 3.27, it is expected that activity and NSS decrease while AIPE increases during the course of the neuropathic disorders. On the other hand, a

typical myopathic disorder exhibits a complete activity, an increase in NSS and a decrease in AIPE.

3.5.2.4.2 Motor Unit Number Estimate (MUNE):

As previously stated, motor unit consists of the spinal alpha motor neuron, its axons, and muscle fibers innervated by the motor neuron. Motor unit number corresponds to the number of anterior horn cells or axons innervating and controlling a single muscle provided that they are intact [98, 99]. In neurological disorders characterized by a loss of lower motor neurons, a patient's force primarily depends upon the number of the functioning motor units in a group of muscles [26]. Motor unit number is a critical measure in any disease involving injury or death of motor neurons or motor axons. Inasmuch it is not possible to count the number of alpha motor neurons in living humans, a motor unit number estimate (MUNE) must serve as a substitute [99]. There is no electrophysiologic technique that allows for simple direct measurement of motor unit number. MUNE may provide information into the underlying disorder, its distribution and severity [98, 100].

The technique is based on the recruitment of successive motor units by gradual intensity of the electrical stimulation to the nerve [10]. Supramaximal stimulation of a peripheral nerve activates all the muscles innervated by this nerve [26]. Then, motor unit number is estimated by comparing an average motor unit parameter with the corresponding parameter of whole muscle [99-101]. The parameter could be twitch or tetanic tension that could provide also information about the contractile properties of the motor units, but it is much more convenient to use parameters in terms of electrical activity [102]. Therefore, amplitudes or "areas" of single motor unit potentials are compared with those of maximum compound action potential (CMAP) of the same muscle. CMAP is also referred as M-wave [101, 103]. Basically, motor unit number

estimate (MUNE) is computed by dividing the CMAP yielded from the supramaximal stimulation of the peripheral nerve by the average of single motor unit potentials (SMUPs) [98][104]. The information obtained through MUNE may enable clinicians to better characterize the extent of motor unit loss and subsequent reorganization of the motor unit pool in response to disorders of the central and peripheral nervous systems [105].

Although all of them are based on the same premise about the comparison of the SMUP to CMAP, a variety of techniques has been described in order to discard the problem of alternation which is referred as the discharge of different axons by the same stimulation. These are, all-or-none increments of CMAP (Manual incremental method), automated incremental method, multiple-point stimulation, F-wave measurements, spike-triggered averaging and statistical estimates (Poisson analysis) [26, 100, 101, 103, 106].

Manual incremental method being the easiest and to most direct approach to counting of SMUPs is the original incremental method based on the all-or-none characteristic of the activation of motor axons. Measurement of successively recruited individual motor units is achieved by the application of finely controlled current in very small steps [26, 98]. The stimulus intensity to a motor nerve is gradually increased above the threshold level and the response that grows in each step is monitored. Each increment is supposed to reflect the excitation of an additional motor unit. At least ten increments are applied and the mean of their peak-to-peak amplitudes are used to compute MUNE by dividing this value into the amplitude of the maximum M-wave for that muscle [100, 104, 107]. The application of manual incremental method is represented in Figure 3.28.

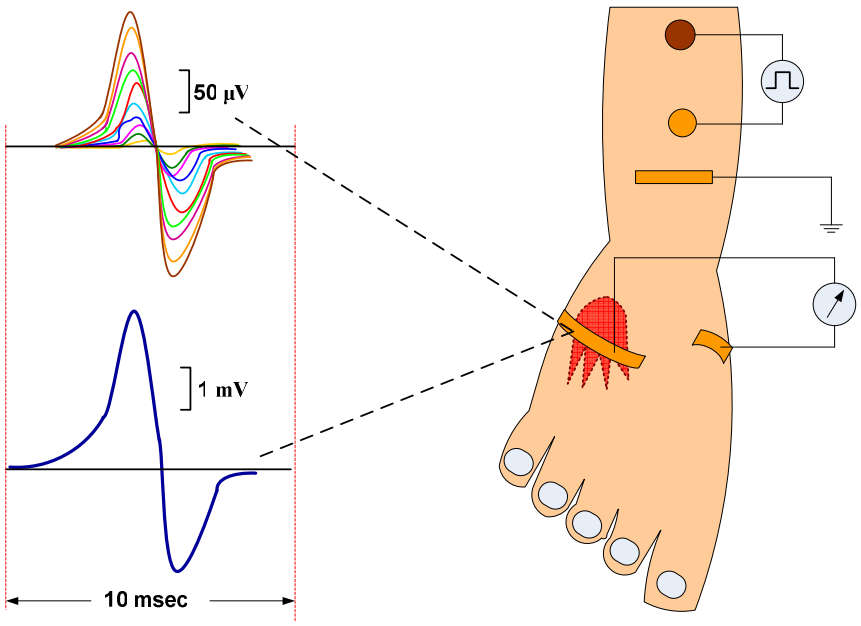


Figure 3.31 Manual incremental method of MUNE. As stimuli of gradually increasing intensity are applied to the peroneal nerve at the ankle, the responses of the extensor digitorum brevis (EDB) muscle grow in steps (top left); each step is assumed to reflect the excitation of an additional motor unit. The mean amplitude of the putative motor unit potential is then divided into the maximum M-wave (bottom left) to give the MUNE.

In automated incremental method, a software program controls the stimulus intensity while the detection of new response increments is achieved by fast Fourier analysis. Individual motor unit potentials are derived by the subtraction of templates in the computer memory and the areas, rather than the amplitudes, of the responses are used in the computations. A separate algorithm detects, and rejects, instances of alternation. The method is quick and any doubtful motor unit potentials can be seen in the montage and corrected for. Largely because of the elimination of alternation, the MUNEs tend to be lower than those obtained by the manual method [100].

F-wave measurement is based on the analysis of F-waves by them treating as the responses of one, or very few, motor units [100, 101]. The F-wave is a late motor

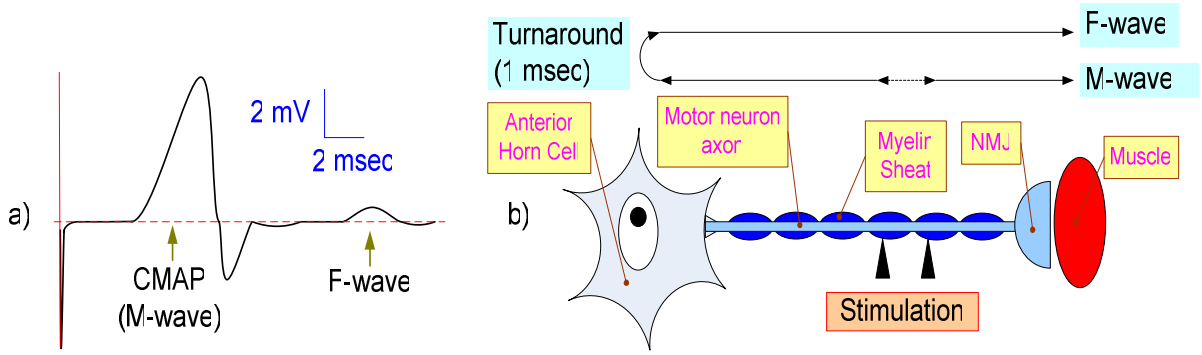


Figure 3.32 a) F-wave following the CMAP as a response of a muscle to nerve stimulation **b)** Schematic representation of the F Response Circuitry.

response that occurs after the compound muscle action potential (CMAP is also known as *M Potential*). In the upper extremity, the F-wave is recorded from the thenar muscles when the median nerve is stimulated at the wrist or from the abductor digiti quinti (ADQ) muscle when the ulnar nerve is stimulated at the elbow. In the lower extremity, the F-wave is recorded from the extensor digitorum brevis (EDB) or from tibialis anterior (TA) when the peroneal nerves is stimulated at the ankle or from abductor hallucis brevis (AHB) or from abductor digiti quinti pedis (ADQP) when tibial nerve is stimulated at the popliteal. The F response is CMAP that represents 1% to 5% of the muscle fibers. The F-wave following the CMAP after the stimulation and the circuitry of this response are illustrated in Figure 3.29 [24, 26].

When motor axons are stimulated, only a small number of neurons will generate F-wave as a recurrent response. This can be used to isolate responses of single motor units [26, 100, 104]. The F-wave analysis is associated with the assumption that the F-wave is a response of single motor unit and that this can be helpful in eliminating the alternation [26, 108].

Multiple point stimulation (MPS) is based on stimulating a motor nerve at different points along its course and accepting only the responses of the one or two motor units with the lowest axonal thresholds [98, 100, 101, 109]. This method is developed in order to eliminate the problem of alternation which results from the activation of multiple motor units [100, 104, 110]. The motor nerve is stimulated at very low intensities just sufficient to elicit a quantal response. Then, the stimulating electrode is moved slightly along the nerve and this process is repeated. The waveform and the amplitude yielded in each stimulation process can be recorded. Afterwards, mean amplitude is calculated and divided into the supramaximal response to compute MUNE [104]. Being a simple and noninvasive technique, multiple point stimulation (MPS), has been shown to be highly reproducible in both normal subjects and patients with ALS [111]. On the other hand, this method is time-consuming and requires skill and training [98, 100].

Unlike other MUNE methods, spike-trigger averaging (STA) technique is performed with the voluntary recruitment [10, 26]. Surface-recorded motor unit potentials are averaging is achieved during relatively weak voluntary contractions [101]. Spike-trigger averaging utilizes two-channel recorder to acquire voluntarily activated motor units as a measure of single motor unit potential. This is the only technique that employs a needle electrode (e.g. single fiber EMG needle) [112]. Single motor units are detected by a needle electrode on the first channel during the weak voluntary contraction [100, 103]. The size of these units is averaged by using a pair of surface electrodes on the second channel [26]. Each discharge of the motor unit triggers an averaging device receiving its input from an electrode over the muscle. In this way the surface-recorded potential of that motor unit can be differentiated from background “noise” generated by other active units. The alternation is eliminated again by means of this technique [100, 104]. Surface potentials are recorded and the single motor unit spike is used as a trigger to time-lock the surface potentials to calculate an average surface response corresponding to the needle recorded spike. Different motor units are recorded by changing needle position. A sample of surface motor unit potentials might be generated and average motor unit amplitude can be computed. Then, MUNE is found by dividing the average single unit response into the

maximum CMAP [104]. Since MUNE techniques is difficult to proximal muscles whose nerves are often inaccessible due to the necessity of using electrical stimuli, STA might be used for this purpose [104]. It is technically more difficult than some other MUNE methods, as it requires a needle electrode insertion and is critically dependent on the operator's ability to trigger only on the intended motor unit. Since it requires needle insertion it is critically dependent on the operator's ability to trigger only on the intended motor. This may be problematic in patients having diseases that may be associated with unstable motor unit. As an example, motor unit morphology may vary from spike to spike in amyotrophic lateral sclerosis (ALS) making difficult the triggering on the same motor unit [104]. Furthermore, MUNE accuracy may be affected by some sources of error with STA method. Volume conduction leading to the contamination of the CMAP by potentials coming from remote muscles and Spurious trigger potentials occuring when the trigger threshold is crossed by either a different voluntary motor unit potential or by an involuntary fasciculation potential are examples for these sources of error [113]. In a previous study, a modified form of STA referred as Decomposition-enhanced spike-triggered averaging (DESTA) was applied to the vastus medialis muscle to examine size distributions of surface-detected motor unit action potentials (S-MUAPs) at various force levels beside to finding MUNE [114].

Statistical MUNE method is based on the relationships between the size of individual components contributing to the CMAP and the random variation in the size of a submaximal CMAP [26, 98]. In this approach, a motor unit is considered as a "quantum" and stimulation of axons with overlapping thresholds will results in different random combinations of quanta. The incidences of the differently sized quantal responses constitute a Poisson distribution [26, 98, 100]. Poisson analysis is applied to multiple submaximal evoked muscle responses via repetitive motor nerve stimulation at different intensities [101, 103]. The identified quantum is then compared with the maximum M-wave [98, 103]. The computation of statistical MUNE is as follows:

$$\sigma^2 = \frac{\left[\frac{\sum x^2 - \left(\sum x\right)^2}{n}\right]}{n-1} \quad (3.1)$$

where σ^2 is the variance, n is the number of M-waves (or CMAP) and x is the individual M-waves.

$$Mean \quad SMUP \quad area = \frac{\text{variance}}{Mean \quad CMAP \quad area - \text{minimum} \quad CMAP \quad area} \quad (3.2)$$

$$MUNE = \frac{\text{maximal} \quad CMAP \quad area}{\text{SMUP} \quad area} \quad (3.3)$$

Statistical MUNE measurement includes three steps. First, the intensity levels that will be used to measure variance must be selected. Then, the variance of a sequence of thirty CMAP is found. The last step is the calculation of an SMUP estimate and MUNE from the variance at each stimulus intensity level tested and the maximal CMAP size. The stimulus intensity is selected by performing a ‘‘scan’’ of the CMAP by applying thirty stimuli of equally graded intensity up to a supramaximal stimulus [98]. Statistical MUNE has some advantages compared with the other commonly used methods. The statistical method can test a larger proportion of the MUP population in a muscle than the other methods and can be applied at any level of stimulus intensity, assuring that the full range of SMUPs in the CMAP is tested. The scan permits identification of segments that need to be tested. Small MUPs that might not be seen with other methods can be identified [98]. There are several drawbacks. The technique must be learned and practiced in order to become sufficiently skilled to obtain reliable results. In addition, some patients can not tolerate the multiple electrical stimuli. An unstable MUAP will result in loss of accuracy of up to 10%. It is easy to overlook small MUPs when not looking for them [98].

Even though they vary due to the different techniques, normal values range from 200 to 350 for the thenar muscles tested by means of the stimulation of the median nerve and from 150 to 220 for the extensor digitorum brevis (EDB) tested with the stimulation of the peroneal nerve [26, 103].

In aging MUNE has shown a progressive loss of functioning motor units, especially after the age of 60 years. There is approximately a decline to half of the MUNE over 60 years of age compared to younger subjects [115]. Among the muscles studied, the extensor digitorum brevis (EDB) has the most prominent loss, and the brachial biceps shows the least change [100, 101, 103]. Using the incremental technique, studies showed little change until age 60 [104].

MUNE have been shown to change as expected not only with normal aging and but also in many neurogenic disorders. MUNE is only applied to limb muscles in neurogenic processes where a good correlation is observed between MUNE and reduced recruitment. MUNE can provide a reliable quantitative measure in axonal neuropathy, radiculopathy or any neurogenic process with axonal loss. Nevertheless, it has been applied primarily in amyotrophic lateral sclerosis (ALS) [98, 116].

Electromyography (EMG) is useful in the diagnosis of amyotrophic lateral sclerosis (ALS) but provides little information for adequate assessment of ALS and its course. Nonetheless, other electrophysiological techniques are available to study quantitatively pathophysiological changes in ALS muscles such as motor unit loss and the size increase of the remaining motor units [117]. Unfortunately, most of these techniques are too sophisticated for routine clinical use [118]. MUNE is reduced in ALS before denervation evidence such as clinical weakness and MUAP alterations is observed [98, 101, 103]. This is due to the some surviving motor neurons which enlarge their territories through axonal sprouting being sufficient to maintain CMAP and twitch tensions within normal

limits [98, 101, 103, 104]. Therefore, since collateral reinnervation may delay decrease in strength, MUNE may be considered as a valuable diagnostic tool in ALS [101]. MUNE may have a role distinct in identifying early involvement by ALS by monitoring decline of motor neurons in the involved region before muscle strength is affected [119]. Besides, MUNE may be also used to predict the patient's survival [119, 120]. As might be anticipated, motor unit estimation has been proven as effective in the diagnosis and assessment of patients with spinal muscular atrophy (SMA), poliomyelitis, peripheral neuropathies and Charcot-Marie-Tooth (CMT) neuropathy [103, 121].

Human incremental MUNE studies have indicated that subjects in myopathic diseases such as Duchenne, limb-girdle and a variety of inflammatory and metabolic myopathies had normal motor unit numbers. However, reduced MUNE was noted in myotonic patients when the incremental technique is performed. Even though this result was surprising initially, subsequent reports have documented a neurogenic component to myotonic dystrophy confirming a reduction in motor unit number [103, 104].

There are also some studies to reveal decrease in the number of motor neurons in some neurological disease such as Parkinson's disease (PD) resulting from the degenerative alterations. MUNE has been used to assess lower motor neuron assessment [122].

MUNE Methods are used also to monitor the viability of motor neuron pools and the muscle fibers at or below the level of spinal injury and to study the long-term effects of the injury [123].

In MUNE techniques it is assumed that each increment in response is due to the excitation of an additional motor unit. However, this assumption may not be always true

because several motor axons might have similar thresholds, and may, behave as a single large unit [103]. In addition, alternation which means the overlap of other axons firing over a given range of stimulus amplitude may result in fictitiously high motor unit estimates [98, 100, 103].

4. NEUROMUSCULAR DISEASES

4.1 Introduction

Electrodiagnostic examinations such as nerve conduction studies (NCSs) and electromyography (EMG) have been used to detect and characterize the disease processes affecting the motor units [10]. These processes are basically referred as the neuromuscular diseases (NMDs). NMDs can be defined as a group of diseases that affect any part of the nerve and muscle. Neuromuscular diseases compromise a very broad range of disorders that impair the functioning of the muscles either directly via intrinsic muscle pathology, or indirectly via nerve pathology [124]. Most of the NMDs cause weakness and wasting of skeletal muscles. The distinguishing features of these diseases vary depending on which of the four functional components of the motor unit such as the cell body of the motor neuron, its axon, the neuromuscular junction or the muscle fibers innervated by the motor neuron is primarily affected [125].

In clinical sense, the disorders yielding from the pronounced changes in nerve cell bodies or peripheral nerves are called neurogenic diseases. On the other hand, the disorders resulted from the degeneration of the muscle are called myopathic diseases [95]. The neuromuscular diseases are classified in the Table 4.1.

4.2 Motor Neuron Diseases (MNDs)

Motor neurons are nerve cells located in the brain, brainstem, and spinal cord that serve as controlling units and vital communication links between the nervous system and the voluntary muscles. Messages from motor neurons in the brain which are called upper

Table 4.1

Classification of neuromuscular disease

<table>
<tr><td colspan="5">Neurogenic Disorders</td></tr>
<tr><td>I) Motor Neuron Disease</td><td colspan="4">II) Peripheral Neuropathies</td></tr>
<tr><td>Amytrophic Lateral Sclerosis (ALS)
Spinal Muscular Atrophy (SMA)
Poliomyelitis and Post-polio syndrome (PPS)</td><td>Polyneuropathies

Neuropathies Associated with General Medical Conditions
Diabetic Neuropathy

Alcoholic Neuropathy
Uremic Neuropathy
Neuropathies in Malignancies
Inflammatory, Infective and Autoimmune Neuropathies
• Guillain-Barré syndrome (GBS)
• Chronic Inflammatory Demyelinating Polyneuropathy (CIDP)
• Multifocal Motor Neuropathy with Conduction Block (MMNCB)
Metabolic and Toxic Polyneuropathies
Inherited Neuropathies
Charcot-Marie-Tooth Disease Type I (HMSN Type I)
Charcot-Marie-Tooth Disease Type II (HMSN Type II)</td><td>Mononeuropathies

Median Mononeuropathy

Ulnar Neuropathy
Radial Mononeuropathy
Femoral Mononeuropathy
Peroneal Mononeuropathy

Tarsal Tunnel Syndrome (TTS)</td><td colspan="2">Radiculopathies and Plexopathies

Cervical Radiculopathy

Brachial plexopathy
Lumbosacral Plexopathy</td></tr>
<tr><td colspan="5">Neuromuscular Disorders</td></tr>
<tr><td colspan="5">Myasthenia gravis (MG)</td></tr>
<tr><td colspan="5">Lambert-Eaton Myasthenic Syndrome (LEMS)</td></tr>
<tr><td colspan="5">Botulism</td></tr>
<tr><td colspan="5">Myopathic</td></tr>
<tr><td>Muscular Dystrophies (MD)</td><td>Metabolic Myopathies</td><td>Myositis</td><td>Endocrine Myopathies</td><td>Congenital myopathy</td></tr>
<tr><td>Duchenne Muscular Dystrophy
Becker Muscular Dystrophy
Facioscapulohumeral Muscular Dystrophy (FSHD)
Limb-girdle Muscular Dystrophy</td><td>Acid Maltase Deficiency (Type II Glycogenosis)
Debrancher Deficiency (Type III Glycogenosis)
Muscle Phosphorylase Deficiency (Type V Glycogenosis)
Muscle Phosphokinase Deficiency (Type VII Glycogenosis)
Disorders of Lipid Metabolisms
Mitochondrial Disease</td><td>Dermatomyositis
Polymyositis
Inclusion Body Myositis</td><td>Adrenal dysfunction
Thyroid dysfunction
Parathyroid dysfunction
Pituitary dysfunction
Islands of Langerhans dysfunction
Steroid myopathy</td><td></td></tr>
</table>

motor neurons are transmitted to motor neurons in the spinal cord being referred as lower motor neurons and then, transmitted to the muscle fibers included in the motor units of particular muscles.

MNDs are degenerative disorders of aging nervous system where upper and lower motor neurons are affective [26]. They are characterized by the gradual degeneration and death of motor neurons. The most prominent MNDs are amyotrophic lateral sclerosis (ALS), spinal muscular atrophy (SMA), poliomyelitis and post-polio syndrome (PPS).

4.2.1 Amytrophic Lateral Sclerosis (ALS):

Amyotrophic lateral sclerosis (ALS), which is also called Lou Gehrig's disease, is a rapidly progressive, invariably fatal neurological disease that attacks the motor neurons responsible for controlling voluntary muscles [26, 120, 126]. Both upper motor neurons (UMN) and lower motor neurons are affected [117]. Although young patients may be affected, it often occurs in individuals being 55 to 60 years old. A slight predominance of males can be noticed [24].

The etiology of the ALS is unclear. Genetic, immunologic, infectious and toxic etiologies have been proposed. However, none of them have been proven [24].

Signs and symptoms of LMN dysfunctions are muscle atrophy, weakness, fasciculations and cramps. UMN dysfunction is observed as stiffness, slowness of movement, spasticity, weakness, patologic hyperreflexia and Babinski responses [24]. The sign and symptoms may wax and wane. An apparent improvement may occur due to the reinnervation and collateral sprouting. However, ALS progresses without remission

and may result in death due to the respiratory failure within 3 to 5 years from the onset of symptoms. However, about 10 percent of ALS patients survive for 10 or more years [24, 26].

Electrophysiological studies such as EMG and NCS are helpful in the diagnosis of ALS. Fibrillation potentials and positive sharp waves are seen due to the diffuse denervation [26, 36]. Fasciculation potentials can also be observed but they are non-specific. These abnormalities exhibit asymmetric distribution, especially in early stages. Large fibrillation potentials may suggest chronic denervation. MUAPs have large-amplitude and polyphasic waveforms [127]. Some MUAPs may have satellite potentials. MUAPs are reduced in number, recruited poorly and discharge rapidly resulting in incomplete interference pattern. Since the motor unit population is suggested to decrease by half in each 6 months period of the first year and then to diminish more slowly thereafter, surviving enlarged motor units contribute less twitch force As a result, they fatigue more easily than the normal motor units due to the mechanical insufficiency [24, 26]. In this context, MUNE may be helpful to predict the disease progression [26]. SFEMG findings are increased fiber density and jitter values as a measure of collateral reinnervation [26, 128]. In nerve conduction studies (NCS), motor fiber conduction velocity is within normal range in nerves of unaffected muscles. However, it is not less than 70 percent of the average value in nerves of severely affected muscles. The conduction velocity of the sensory nerves is normal even in severely affected limbs [24, 26].

4.2.2 Spinal Muscular Atrophy (SMA):

SMA is also a motor neuron disease which results in selective degeneration of the lower motor neurons. SMA is currently classified as Type I, II and III. They are also

known as SMA I (Werdnig-Hoffmann disease), SMA II (Juvenile SMA) or SMA III (Kugelberg-Welander disease) [129].

SMA has an autosomal recessive trait with deletion of the survival motor neuron (SMN) gene on chromosome 5q13 in more than 90 percent of infantile cases [24, 26].

SMA I (Werdnig-Hoffmann disease) which is also known as infantile SMA is the most severe type. The onset of this type is the first month of the life and usually results in death before age 2 years. Its clinical features comprise progressive muscle weakness, atrophy of the trunk and extremities, hypotonia and feeding difficulties [24, 26]. About half of the patients have fasciculations in the tongue but they can be observed less frequently in the atrophic muscles [26]. In EMG studies, fibrillation potentials and positive sharp discharges may be seen but their incidence depends on stage, progression and severity of the disease. Fasciculation potentials occur rarely. Delayed recruitment of MUAPs demonstrates the loss of anterior horn cells. High-amplitude and long-duration MUAPs are seen as expected due to the collateral sprouting and high fiber density [24, 26, 36]. Incomplete interference pattern is produced by maximal voluntary contraction [26]. Nerve conduction studies for motor neurons indicate nearly normal velocities beside a CMAP with reduced amplitude. Sensory nerve studies usually reveal normal amplitudes and velocities [24, 26].

SMA II and SMA III (Kugelberg-Welander disease) are both included into the category of juvenile SMA. They appear in early childhood or during adolescence or adulthood and have better prognosis. The clinical characteristics seen at the beginning are the proximal muscle weakness and atrophy in the lower limbs. The symptoms are seen initially in the extensor muscles of the hip and knees. Subsequently, the involvement of the shoulder girdle muscles occurs. One half of the cases have fasciculations in the proximal muscles [26, 36]. In EMG examinations, fibrillation potentials may be seen with

higher percentage especially in more severely affected patients. Fasciculations may be observed. If complex repetitive discharges are detected it should suggest a late stage. All of these spontaneous activities are observed in the lower limbs more than upper limbs and in proximal muscles more than distal muscles. High-amplitude, long-duration MUAPs are detected during the voluntary contraction [36]. These are poorly recruited at maximal effort. Satellite potentials demonstrate the presence of the slow-conducting regenerating axons. In advanced cases, small polyphasic potentials may suggest secondary myopathic alterations of the atrophic muscles. Even though they are usually normal, motor and sensory nerve conduction studies may reveal CMAP moderately reduced in amplitude [10, 24, 26].

Another type of SMA is X-linked Bulbospinal Muscular Atrophy which is also referred as Kennedy' Disease). It affects only men and usually begins between the third and fifth decades of life after a slow progression [24]. The clinical manifestations are mild facial weakness, facial fasciculations being most prominent around the mouth and chin, severe atrophy of the tongue, dysarthria and dysphagia associated with atrophy and weakness of facial, jaw and glossal muscles, postural hand tremor and hyporeflexia [24, 26]. EMG indicates increased insertional activity, fibrillation potentials, complex repetitive discharges (CRDs) in facial muscles, reduced recruitment of large prolonged-duration, polyphasic MUAPs. Nerve conduction studies (NCS) demonstrate absent or low-amplitude sensory nerve action potential (SNAP) despite clinically normal sensation. This is associated with degeneration of the dorsal ganglia. Motor studies reveal normal findings but CMAP amplitude may be low if recorded from weak and wasted muscles [10, 24, 26].

4.2.3 Poliomyelitis and Post-polio syndrome (PPS):

Poliomyelitis is a paralytic disease causing degenerative changes in anterior horn cells and brainstem as a result of inflammatory reaction in the meninges. The etiologic agent is polio virus [24, 26]. Symptoms such as fever, headache, myalgia and gastrointestinal disturbance are present in patients with acute polio [24]. Systemic infection may exhibit paralytic illness following the prodromal symptoms. Weakness, wasting and depressed reflexes appear during the first or second week of the illness. Weakness is asymmetric and lower extremities are most commonly involved [24, 26]. The sensory system is normal in the neurologic examination [26]. Increased fiber density, jitter and macro MUPs demonstrate pronounced and unstable reinnervation as compensation for the loss of motor neurons [26]. EMG initially shows reduced recruitment patterns during the acute phase of poliomyelitis. As the degeneration of motor axons occurs, fibrillation potentials can be observed. Subsequent reinnervation results in diminution of spontaneous of spontaneous discharges and motor unit potentials of large amplitude and long duration begin to appear [36]. In nerve conduction studies, normal velocities with reduced amplitude of CMAP are revealed in proportion to the degree of muscle atrophy [26].

Post-polio Syndrome can reveal in cases that had poliomyelitis previously and recovered with least or mild sequela. Muscle wasting and loss of muscular function appear due to the impairment of the anterior horn cells that have remained intact in the previous infection [24].

4.3 Peripheral Neuropaties

Disorders of peripheral nerves cause to not only sensory symptoms occurring spontaneously without an external stimulus such as numbness, tingling etc but also motor

signs such as weakness being proximal in acute case and which is distal in chronic disorders. Peripheral neuropathies can be classified as polyneuropathies, mononeuropathies and radiculopathies and plexopathies.

4.3.1 Polyneuropathies:

Polyneuropathy is a neurological disorder that occurs when many peripheral nerves throughout the body malfunction simultaneously. The temporal course of a polyneuropathy may be acute, subacute, chronic, progressive, stepwise, relapsing/remitting fashion. The nerve fibers involved in a polyneuropathy can be categorized as motor fibers, large sensory fibers, small sensory fibers or autonomic fibers. The pattern of the polyneuropathy can be subdivided as distal dying back (distal-to-proximal gradient), short nerve involvement, symmetric an asymmetric. The underlying pathologic processes of polyneuropathies can be separated as axonal loss or demyelination.

4.3.1.1 Neuropathies Associated with General Medical Conditions:

Since they accompany diverse clinical conditions, neuropathies associated with general medical conditions are the most commonly encountered polyneuropathies in clinical practice. These are diabetic neuropathy, alcoholic neuropathy, uremic neuropathy, neuropathies in malignant conditions, neuropathies associated with paraproteinemia, necrotizing angioplasty, sarcoid neuropathy, Sjörgen's Syndrome.

4.3.1.1.1 Diabetic Neuropathy

Diabetic neuropathies are neuropathic disorders that are associated with diabetes mellitus. These conditions are suggested to result from diabetic microvascular injury involving small blood vessels that supply nerves called vasa nervorum. They are characterized by the demyelination secondary to axonal loss [26].

Patients with signs of neuropathy have slower nerve conduction velocities and smaller amplitudes than those without symptoms. This demonstrates a correlation between clinical findings and the degree of conduction changes [26]. Electromyographic sampling of distal lower extremity muscles may reveal acute and ongoing denervation in the form of positive sharp waves and fibrillation potentials. Reinnervation changes such as large-amplitude, long-duration, polyphasic motor unit potentials reflect chronicity. Single-fiber studies may indicate a measure of reinnervation [26].

4.3.1.1.2 Alcoholic Neuropathy

In most cases of alcoholic neuropathy, the onset of the polyneuropathy is insidious and prolonged, but in some cases, it can appear acutely over a period of few days. Severe cases of alcoholic neuropathy can lead to the development of symptoms in the proximal lower extremities and distal upper extremities. Initially, sensory complaints such as distal pain, paresthesias and dysesthesias occur first in the leg and then, in the arms. In more advanced cases, bilateral foot-drop associated with distal muscular atrophy involving extensor rather than the flexors.

Electrophysiologic examinations show impaired function of small caliber motor fibers and large cutaneous sensory fibers. Nerve conduction studies reveal initially normal or slightly reduced velocities in most patients. Velocity decreases as the loss of evoked sensory or motor responses progress [26]. Needle EMG is based on presentation. A typical peripheral neuropathy screen will involve a proximal muscle and a distal one in the lower and upper extremities. Significant abnormalities seen in patients with alcoholic neuropathy include the presence of positive sharp waves and/or fibrillation potentials. Complex, repetitive discharges also may be observed. However, if the NCSs are normal, the presence of positive sharp waves in one intrinsic foot muscle is not necessarily indicative of neuropathic pathology [26].

4.3.1.1.3 Uremic Neuropathy

Uremic neuropathy is a distal sensorimotor polyneuropathy caused by uremic toxins. The severity of neuropathy is correlated strongly with the severity of the renal insufficiency. It often develops in patients with chronic renal failure or in patients that undergo chronic hemodialysis. Uremic neuropathy is considered a dying-back neuropathy or central-peripheral axonopathy associated with secondary demyelination [26].

Sensory and motor conduction abnormalities are seen in all limbs in patients with severe renal insufficiency. Greater deficits in the peroneal nerve are more common than the median nerve. Prolonged F wave latencies of tibial and peroneal nerves are the profound and reproducible abnormalities in patients with chronic renal failure. Both sensory and motor nerve conduction velocities are reduced. Reduced compound action potential amplitudes are due mainly to reduced densities of large myelinated motor and sensory fibers. EMG reveals minimal or absent fibrillation or positive sharp wave. Only more advanced cases of uremic neuropathy lead to predominantly distal muscle denervation [26].

4.3.1.1.4 Neuropathies in Malignancies

Peripheral nerves can be affected directly or indirectly in malignant conditions. Lymphomas and leukemias may infiltrate the peripheral nerves through hematogenous spread. On the other hand, non-lymphomatous solid tumors may lead to external compression. Almost one third of the patients with malignancies develop clinically latent neuropathies.

Nerve conduction studies show abnormalities involving both large and small fibers. Substantial reduction in amplitude of sensory nerve action potential (SNAP) can be observed in addition to mild slowing of sensory and motor fibers. EMG shows fibrillation potentials and high-amplitude and long duration MUAPs [26].

4.3.1.2 Inflammatory, Infective and Autoimmune Neuropathies

These polyneuropathies occur as a result of damage in peripheral nerves either due to an inflammatory reaction or due to an autoimmune disorder.

4.3.1.2.1 Guillain-Barré syndrome (GBS)

Guillain-Barré syndrome (GBS) is an acute inflammatory demyelinating polyneuropathy (AIDP), an autoimmune disease affecting the peripheral nervous system, usually triggered by an acute infectious process). It is frequently severe and usually exhibits as an ascending paralysis noted by weakness in the legs that spreads to the upper limbs and the face along with complete loss of deep tendon reflexes. Guillain-Barré

syndrome is due to an immune response to foreign antigens (such as infectious agents or vaccines) but mistargeted to host nerve tissues. The targets of such immune attack are thought to be gangliosides, which are complex glycosphingolipids present in large quantities on human nerve tissues, especially in the nodes of Ranvier. The end result of such autoimmune attack on the peripheral nerves is inflammation of myelin and conduction block, leading to a muscle paralysis that may be accompanied by sensory or autonomic disturbances [24, 26].

During the first days of the illness, all nerve conduction studies may be normal. Later, motor nerve conduction studies show prolonged distal latencies with conduction block and temporal dispersion as the other evidence of segmental demyelination [24]. The needle EMG indicates the demyelinating pattern such absence of denervation potential, normal MUAP morphology, but reduced recruitment in weak muscles. In early stages of the disease, there is no spontaneous activity at rest. However, myokymic discharges can be observed in the limbs and in facial muscles. After the denervation, MUAPs may become more polyphasic, subsequently an increase in amplitude and duration can be seen. A reduced interference pattern can be shown in EMG [24, 26, 130].

4.3.1.2.2 Chronic Inflammatory Demyelinating Polyneuropathy (CIDP)

CIDP is acquired demyelinating motor and sensory neuropathy assumed as immun-mediated disease. Both proximal and distal muscles are affected. It is a progressive disease. It is characterized by areflexia or hyporeflexia. Large-fiber sensory loss such as touch, vibration, position sense is more common to small-fiber sensory loss (e.g. pain, temperature) [24].

In nerve conduction studies, evidence of primary demyelination such as markedly prolonged distal latencies particularly in upper limbs, markedly slowed conduction velocities are present. Distal CMAP and SNAP amplitudes are reduced particularly in lower limbs. Needle EMG indicates evidence of chronic and ongoing axonal loss such as fibrillation potentials, MUAP with large amplitude and long duration having reduced recruitment [10, 24, 26, 107].

4.3.1.2.3 Multifocal Motor Neuropathy with Conduction Block (MMNCB)

It is a pure motor neuropathy associated with antiganglioside antibodies (e.g. anti GM_1). The patients demonstrate a pure lower motor neuron syndrome which is similar to amytophic lateral sclerosis (ALS) [24, 26]. However, evidence of acquired segmental demyelinating neuropathy with conduction block along motor nerves is found in electrophysiologic studies [24, 131].

Findings are similar to CIDP in motor nerve conduction studies. There is evidence of demyelination such as prolonged latencies, slowed conduction velocities and prolonged late responses. Needle EMG findings typically indicate decreased recruitment of MUAPs in weak muscles as a result of proximal conduction blocks [24, 131].

4.3.1.3 Metabolic and Toxic Polyneuropathies

These neuropathies yield from either nutritional disturbances or toxic causes. Neuropathies are due to the specific nutritional deficiencies such as beriberi, pellagra and pernicious anemia. Toxic neuropathies occur after the administration of several drugs or the exposure to chemical substances such as lead or arsenic [26].

In nutritional neuropathies, diets deficient in vitamins and other nutritional factors have a major role in the polyneuropathies associated with beriberi, pellagra, pernicious anemia, dysentery and cachexia. They lead axonal degeneration. Diffuse spontaneous discharges are detected in EMG studies especially in patients with prominent axonal degeneration. However, motor nerve conduction studies are observed as normal [26].

In toxic neuropathies, three sites of cellular involvement such as neuropathies affecting cell bodies particularly those of the dorsal ganglion, myelinopathy or Schwannopathy with primary segmental demyelination and distal axonapathy that cause dying-back axonal degeneration [26]. A variety of drugs such as allopurinol, chloramphenicol, cis-platine and industrial chemicals such as acrylamide, inorganic mercury, vinyl chloride cause distal axonapathy [26]. EMG studies show fibrillation potentials and positive sharp waves. In addition, compound nerve and muscle action potentials in reduced amplitude can be seen [26].

4.3.1.4 Inherited Neuropathies

Hereditary motor and sensory neuropathy (HMSN) comprises several types such as hypertrophic and neuronal varieties of Charcot-Marie-Tooth (CMT) disease, Dejerine-Sottas disease, Refsum disease.

HMSN can also have genetic classification. CMT1 is the demyelinative or hypertrophic form and is an autosomal dominant disease genetically localized on chromosome 17 (CMT1A) or chromosome 1 (CMT1B). The third form is CMT1C. The loci where this form is mapped are unknown. These are included into HMSN Type I. There are also three different forms of neuronal types (CMT2) mapping to chromosome 1p, 3q and 7p. These forms are CMT2A, CMT2B, CMT2C and are included into HMSN

Type II. X-linked HMSN or CMTX type has three forms such as CMTX1 (X-linked dominant), CMTX2 and CMTX3 (X-linked recessive patterns) [26].

4.3.1.4.1 Charcot-Marie-Tooth Disease Type I (HMSN Type I)

CMT type 1 (CMT1) is primarily a demyelinating neuropathy, anatomic changes directly affect the myelin sheath, with secondary axonal changes. In areas of focal demyelination, impulse conduction from one node of Ranvier to the next is slowed as current leakage occurs and the time for impulses to reach threshold at successive nodes of Ranvier is prolonged, producing slowing of conduction velocity along the nerve segment. In CMT1, atrophy involves peroneal muscles. Nerve conduction studies show a marked, diffuse and uniform slowing [26].

4.3.1.4.2 Charcot-Marie-Tooth Disease Type II (HMSN Type II)

CMT type 2 (CMT2) is a primary axonal neuropathy with changes in both the axon and the nerve cell body. CMT2 tends to affect the lower extremities more than the upper extremities. CMT2 is often a clinically less severe disease than CMT1. Patients with CMT2 may have lower extremity involvement, although clinically they are not easily distinguished from patients with CMT1. Previous studies have shown that no significant side-to-side difference exists in nerve conduction abnormalities or strength, and, like CMT1, the sensory deficit is usually less severe than the motor deficit. In electrophysiologic studies mild slowing of nerve conduction velocities accompanying to compound sensory and motor action with reduced amplitude are observed [26].

4.3.1.4.3 Charcot-Marie-Tooth Disease X-Linked Dominant Type (X-Linked HMSN)

CMT X is an X-linked dominant, primarily demyelinating neuropathy with a mutation in the connexin 32 gene (*CX32*) that codes for a membrane protein (gap junction protein, beta 1) involved in the formation of gap junctions. CMT X1 is clearly a distinct entity. Some varieties of CMT X1 may exhibit abnormal temporal dispersion and heterogeneous conduction velocities that are very atypical of other hereditary neuropathies [26].

Electrophysiologic studies indicate a substantial loss of distal motor and sensory nerve fibers with primary axonal degeneration, a non-uniform slowing of motor conduction velocities and dispersion of CMAP similar to acquired chronic demyelination [26].

4.3.1.4.4 Dejerine-Sottas Disease (HMSN Type III)

Point mutations in the PMP22 or the myelin protein zero gene (MPZ) may cause Dejerine-Sottas disease. Thus, many cases of Dejerine-Sottas disease are now considered severe phenotypes within the genotypic spectrum of CMT1. Congenital hypomyelinating sensory and motor neuropathy is a severe and often fatal newborn disorder that presents with respiratory distress at birth and has been linked to the early growth response gene 2 (EGR2) in some families [26].

Nerve conduction studies demonstrate marked slowing of the motor and sensory fibers. EMG shows focal axonal loss with evidence of severe denervation which is restricted to the territory of the affected nerve [26].

4.3.2 Mononeuropathies

Mononeuropathy is a type of neuropathy that only affects a single nerve. Physical injury is the most common cause of a mononeuropathy. Injury is commonly caused by the prolonged pressure on a nerve that runs close to the surface of the body near a prominent bone, such as a nerve in an elbow, a shoulder, a wrist, or a knee. They are also referred as entrapment monopathies [26].

4.3.2.1 Median Mononeuropathy

Carpal tunnel syndrome (CTS) is the most common entrapment neuropathy in the upper extremity. The median nerve crosses from the distal forearm to the hand through the carpal tunnel. The floor of the carpal tunnel is formed by the carpal bones and the roof by the transverse carpal ligament. Compression of the median nerve by the transverse carpal ligament (flexor retinaculum) can occur. The condition is usually bilateral, although the dominant hand tends to be more severely affected [24, 26].

In nerve conduction studies distal CMAP and SNAP amplitudes generated by stimulating the median nerve at the wrist will be decreased provided that either demyelination with conduction block or axonal loss is present. A markedly prolonged distal motor latency may be observed by stimulating the median nerve at the wrist and recording the response at the abductor pollicis brevis (APB) muscle. However, in over

50% of patients with CTS, distal median motor latency is within the normal limit [24, 26]. EMG studies show fibrillation potentials and positive sharp waves in the intrinsic hand muscles innervated by median nerve in advanced cases [24, 26].

4.3.2.2 Ulnar Neuropathy

The ulnar nerve is an extension of the medial cord of the brachial plexus. This is a mixed nerve that supplies innervation to muscles in the forearm and hand and provides sensation over the medial half of the fourth and the entire fifth digit of the hand, the ulnar part of the palm, and the ulnar portion of the posterior aspect of the hand (dorsal ulnar cutaneous distribution). The most common site of entrapment is at or near the elbow region, especially in either the region of the cubital tunnel or the ulnar groove. The second most likely location of entrapment is at or near the wrist, especially in the area of the anatomic structure called Guyon canal. However, entrapment can occur in the forearm between these 2 regions, below the wrist within the hand, or above the elbow [18, 24].

Ulnar neuropathy occurs as a result of chronic mechanical compression or stretch, either at the groove or at the cubital tunnel at the elbow. The most common cause of this mononeuropathy is the external compression and repeated trauma. There are two entrapments at the elbow; tardy ulnar palsy and cubital tunnel syndrome. The former results form the antecedent traumatic joint deformity or recurrent subluxation. The latter occurs due to the entrapment of the ulnar nerve under humeral-ulnar aponeurosis (HUA). This is due to the repeated flexion of the elbow [24, 26].

Ulnar neuropathy in Guyon's canal through where the ulnar nerve enters the hand at the wrist may occur less commonly. The entrapment is associated with the trauma and

wrist fracture. However, it can be due to a ganglion cyst within the Guyon's canal compressing the ulnar nerve [24, 26].

Involvement of palmar branch containing the motor branch of ulnar nerve may be due to the repetitive movement or pressure of the wrist as in case of bikers [24, 26].

Nerve conduction studies used to measure basic sensory and motor nerve parameters such as latency, amplitude, and conduction velocity. Electrodes (metallic reusable or pregelled disposable tape) are placed over the main belly of the muscle (active) such as the abductor digitorum quinti (ADQ) or first dorsal interosseous (FDI) and the tendon of the fifth or first digit, respectively. The ulnar nerve is stimulated at the wrist and above and below the elbow. This helps localize the site of involvement. Needle EMG examination is helpful in the evaluation of motor unit morphology and recruitment patterns. It ascertains ongoing loss of muscle fibers via detection of abnormal spontaneous activity such as fibrillation potentials and fasciculations. It can be used to check the integrity of the muscle membrane to expand differential diagnosis (eg, myotonia, paramyotonia, periodic paralysis) as manifested by increased insertional activity such as complex repetitive discharges, myokymia, and myotonic discharges [24, 26].

4.3.2.3 Radial Mononeuropathy

Radial neuropathies can be classified as lesions at the spiral groove, lesions in the axilla, and posterior interosseous neuropathy. Since radial nerve passes over the humerus, it is susceptible to compression and hence radial neuropathy at the spiral groove is the most common radial mononeuropathy. It can occur following prolonged immobilization. Radial neuropathy in the axilla occurs when the crutches are used inappropriately due to

the compression of the radial nerve at the axilla. Weakness of arm extension (triceps brachii) and sensory disturbance of the posterior forearm and arm can accompany originating from the disturbance of the posterior cutaneous nerves of the forearm and arm. In posterior interosseous neuropathy (PIN) wristdrop and fingerdrop occur [24].

The most important nerve conduction study in this case, is radial motor study. Radial CMAP can be recorded over extensor indicis poroprius (EIP) muscle following the stimulation of the radial nerve in the forearm at the elbow. The recorded CMAP should be compared to the normal value of 2 to 5 mV. Any axonal loss will result in decreased distal CMAP amplitude after 3 to 5 days. EMG results will be normal in weak muscles with decreased activation of normal MUAPs [24].

4.3.2.4 Femoral Mononeuropathy

Femoral neuropathies can occur secondary to direct trauma, compression, stretch injury, or ischemia. Femoral neuropathy causes weakness predominantly of the quadriceps. Most cases of femoral mononeuropathy yield from the positioning and compression during the pelvic and abdominal surgery [24].

The femoral nerve is part of the lumbar plexus. It is formed by L2-4 roots and reaches the front of the leg by penetrating the psoas muscle before it exits the pelvis by passing beneath the medial inguinal ligament to enter the femoral triangle just lateral to the femoral artery and vein. Approximately 4 cm proximal to passing beneath the inguinal ligament, the femoral nerve is covered by a tight fascia at the iliopsoas groove. The nerve can be compressed anywhere along its course, but it is particularly susceptible within the body of the psoas muscle, at the iliopsoas groove, and at the inguinal ligament [24].

The main motor component innervates the iliopsoas (a hip flexor) and the quadriceps (a knee extensor). The motor branch to the iliopsoas originates in the pelvis proximal to the inguinal ligament. The sensory branch of the femoral nerve, the saphenous nerve, innervates skin of the medial thigh and the anterior and medial aspects of the calf [24].

To detect the femoral mononeuropathy, surface recording electrodes are placed over the one of the quadriceps muscles particularly on the rectus femoris and the femoral nerve is stimulated over below the inguinal ligament. Increased femoral nerve latency and reduced amplitude in CMAP can be observed. In EMG studies, quadriceps muscles demonstrate neurogenic changes such as large-amplitude and long-duration MUAPs [24] [26].

4.3.2.5 Peroneal Mononeuropathy

Peroneal neuropathy is the most common mononeuropathy in the lower limbs. It often occurs at the fibular neck where the nerve is superficical and vulnerable to injury. Peroneal neuropathy can be seen as a result of causes such as trauma, stretch injury or compression due to the prolonged immobilization [24].

Nerve conductions should show isolated peroneal nerve abnormalities. If the lesion is at the knee, then conduction block or, less commonly, conduction velocity slowing over that segment of the nerve should be documented. When axonal loss occurs in direct nerve trauma or with long-standing compression, a small compound muscle action potential may be noted. If other mononeuropathies with conduction blocks are found, then consideration should be made for an underlying vasculitis causing mononeuritis multiplex or possibly for hereditary neuropathy with liability to pressure palsy. If more diffuse nerve abnormalities are noted, then a generalized neuropathy should be considered, especially chronic demyelinating polyneuropathy [24].

EMG is useful to localize the lesion. It can be helpful in determining which nerve is involved primarily—the common peroneal nerve at the knee or one of its two branches, the superficial or deep peroneal nerve. The tibialis anterior or extensor hallicus longus muscles (ie, innervated by the deep peroneal) and the peroneus longus or brevis muscles (ie, innervated by the superficial peroneal) are useful to study for this purpose. Tibialis anterior (TA), extensor hallucis longus (EHL) and peroneus longus (PL) should be examined. MUAP morphology will be normal however decreased MUAP recruitment will be observed [24].

4.3.2.6 Tarsal Tunnel Syndrome (TTS)

Tarsal tunnel syndrome is a condition that is caused by compression of the tibial nerve or its associated branches as the nerve passes underneath the flexor retinaculum at the level of the ankle or distally.

Tarsal tunnel syndrome is a multifaceted compression neuropathy that typically manifests with pain and paresthesias that radiate from the medial ankle distally and, occasionally, proximally. These findings may have a variety of causes, which can be categorized as extrinsic, intrinsic, or tensioning factors in the development of signs and symptoms of tarsal tunnel syndrome. Extrinsic causes may contribute to the development of tarsal tunnel syndrome. Examples include external trauma due to crush injury, stretch injury, fractures, dislocations of the ankle and hind foot, and severe ankle sprains. Local causes may be intrinsic causes of the neuropathy. Examples include space-occupying masses, localized tumors, bony prominences, and a venous plexus within the tarsal canal [24, 26].

Nerve conduction studies should include bilateral tibial (media and lateral plantar) distal motor latencies to both abductor hallucis brevis (AHB) and abductor digiti quinti pedis (ADQP) muscles. This is achieved by stimulating tibial nerve proximal to the tarsal tunnel at the medial malleous. Prolonged distal motor latency are observed and terminal latencies of the abductor hallucis muscle (medial plantar nerve) longer than 6.2 ms are accepted as abnormal. In EMG studies, Fibrillations in the abductor hallucis muscle may be present [24].

4.3.3 Radiculopathies and Plexopathies

4.3.3.1 Cervical Radiculopathy

Cervical radiculopathy is a dysfunction of a nerve root of the cervical spine. The seventh (C7) and sixth (C6) cervical nerve roots are the most commonly affected. In the younger population, cervical radiculopathy is a result of a disc herniation or an acute injury causing foraminal impingement of an exiting nerve. Disc herniation accounts for 20-25% of the cases of cervical radiculopathy. In the older patient, cervical radiculopathy is often a result of foraminal narrowing from osteophyte formation, decreased disc height, degenerative changes of the uncovertebral joints anteriorly and of the facet joints posteriorly [24].

The nerve conduction studies are performed by placing surface electrodes over a muscle belly or sensory area and stimulating the nerve that supplies either the muscle or sensory area from fixed points along the nerve. From this, the amplitude, distal latency, and conduction velocity can be measured. The amplitude reflects the number of intact axons, whereas the distal latency and conduction velocity is more of a reflection of the degree of myelination. [24]

Needle EMG examination involves inserting a fine-needle electrode into a muscle. Electrical activity is generated by the needle insertion into the muscle, voluntary muscle contraction, and the spontaneous firing of motor units. The activity is observed on an oscilloscope screen and quantified; an audible sound is also generated. Denervated muscle produces spontaneous electrical activity while the muscle is at rest. These potentials are called fibrillations or positive sharp waves based on their characteristic shape and sound. Changes can be also seen in the configuration of the individual motor unit, as well as an increase in the firing rate of the individual motor units. The timing of the EMG evaluation is important because positive sharp waves and fibrillation potentials first occur 18-21 days after the onset of a radiculopathy; therefore, it is best to delay this study until 3 weeks after an injury, so that it can be as precise a study as possible [24].

4.3.3.2 Brachial plexopathy

Brachial plexopathy is decreased movement or sensation in the arm and shoulder, caused by impaired function of the brachial plexus (a bundle of nerves that control sensation and movement of the arm). Damage to the brachial plexus is usually related to direct trauma to the nerve, stretch injury, presure from tumors in the area of the brachial plexus, or damage that results from radiation therapy (therapy for some forms of cancer, such as lung cancer). It may be related to congenital abnormalities that cause pressure on the cervical (neck) ribs and may also sometimes be associated with exposure to toxins, chemicals, or drugs [24, 26].

Electrodiagnosis has become a mainstay in the diagnostic evaluation of brachial plexopathies. Electrodiagnostic tests provide physiologic data about the continuity of pathways and of lesion type and severity. Serial testing is helpful to determine prognosis. While positive waves and fibrillations (which indicate axonal injury) do not appear for several weeks after injury, sensory nerve action potentials (SNAPs) can be useful within

days of injury to distinguish a presynaptic lesion from a postsynaptic lesion. With postsynaptic lesions, SNAPs are absent, whereas they are present with presynaptic ganglionic lesions [24, 26, 36].

4.3.3.3 Lumbosacral Plexopathy

Lumbosacral plexus involvement occurs most commonly due to intra-abdominal tumor extension; it occurs less commonly with growth from metastases, lymph nodes, or bone structures. A tumor can invade the plexus directly or track along the connective tissue or epineurium of nerve trunks [24, 26].

The lower (sacral) plexus is involved most frequently (approximately 50%), followed by upper plexus involvement (more than 30%) and panplexopathy (18%). Bilateral plexopathy occurs in 25% of cases and is usually caused by breast cancer r metastases. Lower plexus involvement occurs generally with colorectal and cervical neoplasms. Involvement of the sacral sympathetic nerves is less common (10%) [24, 26].

Electrodiagnostic examinations (electromyography and nerve conduction studies) reveal abnormalities in almost all patients with neoplastic lumbosacral plexopathy. Typical changes include acute and chronic denervation of the lumbosacral plexus. The findings are observed more extensively than would be suspected clinically. Side-to-side comparisons are helpful. Myokymic discharges are not observed. In the segments involved, decreased amplitudes of the evoked motor responses with normal or borderline nerve conduction velocities are noted [24, 26].

4.4 Neuromuscular Junction (NMJ) Disorders

Since neuromuscular junction tries to bridge the gap between the nerve and muscle, transmission of the signal to contract a muscle is blocked with these diseases [132]. The most common of these diseases are myasthenia gravis being an autoimmune disease, Lambert-Eaton Myasthenic Syndrome (LEMS) and Botulism [26].

4.4.1 Myasthenia gravis (MG)

Myasthenia gravis is a chronic autoimmune neuromuscular disease characterized by varying degrees of weakness of the skeletal (voluntary) muscles of the body and fatiguability. It is caused by the circulating antibodies that block acetylcholine receptors at the post-synaptic neuromuscular junction [24, 26].

Muscles become progressively weaker during periods of activity and improve after periods of rest. Muscles that control eye and eyelid movement, facial expression, chewing, talking, and swallowing are especially susceptible. The muscles that control breathing and neck and limb movements can also be affected. In most cases, the first noticeable symptom is weakness of the eye muscles. In others, difficulty in swallowing and slurred speech may be the first signs. Symptoms, which vary in type and severity, may include asymmetrical ptosis (a drooping of one or both eyelids), diplopia (double vision) due to weakness of the muscles that control eye movements, unstable or waddling gait, weakness in arms, hands, fingers, legs, and neck, a change in facial expression, dysphagia (difficulty in swallowing), shortness of breath and dysarthria (impaired speech, often nasal due to weakness of the velar muscles) [24, 26].

In routine motor and sensory nerve conduction studies, CMAP is usually normal being an expected finding in MG. In repetitive nerve stimulation (RNS) studies at 2-3 Hz, muscle action potentials show a decremental response [133]. Since they can also a decremental CMAP response, routine needle EMG should be applied to eliminate denervating disorders such as motor neuron diseases, polyneuropathy, inflammatory neuropahty to distal and proximal muscles especially weak muscles. Patients with moderate to severe myastenia gravis may demonstrate unstable or short, polyphasic MUAPs with normal recruitment. If all of these electrophysiologic studies are normal in patients suspected of having MG, single fiber EMG (SFEMG) should be performed in extensor digitorum communis (EDC) muscle to investigate jitter and blocking. Normal SFEMG findings in a clinically weak muscle discard an NMJ disorder [24, 26, 38].

4.4.2 Lambert-Eaton Myasthenic Syndrome (LEMS)

Lambert-Eaton myasthenic syndrome (LEMS) is a rare disorder of neuromuscular transmission. It is a presynaptic disorder of neuromuscular transmission in which quantal release of acetylcholine (ACh) is impaired, causing a unique set of clinical characteristics, which include proximal muscle weakness, depressed tendon reflexes, posttetanic potentiation, and autonomic changes. The initial presentation can be similar to that of myasthenia gravis, but the progressions of the two diseases have some important differences [24].

LEMS results from an autoimmune attack directed against the voltage-gated calcium channels (VGCCs) on the presynaptic motor nerve terminal. This results in a loss of functional VGCCs at the motor nerve terminals. The number of quanta released by a nerve impulse is diminished. However, because presynaptic stores of ACh and the postsynaptic response to ACh remain intact, rapid repetitive stimulation or voluntary activation that aids in the release of quanta will raise the endplate potential above

threshold and permit generation of muscle action potential. As neuromuscular transmission is completed at additional neuromuscular junctions, a transient increase will occur in the strength of the muscle. Parasympathetic, sympathetic, and enteric neurons are all affected. Clinically, this phenomenon is noted by the appearance of previously absent tendon reflexes following a short period of strong muscle contraction by the patient [24, 26].

While LEMS may be found as a solitary disease, 50% of cases have an associated malignancy. Malignancies that may be found with LEMS may include small-cell lung cancer, lymphoma, non-Hodgkin's lymphoma, T-cell leukemia, non-small lung cancer, prostate cancer, and transitional cell carcinoma of the bladder [26].

Electrophysiologic evaluation of LEMS is important in diagnosis. Routine motor and sensory nerve conduction studies should be performed at least two nerves, a motor and sensory nerve in one upper and one lower extremity. CMAP amplitudes are diffusely low with normal latencies and conduction velocities. Repetitive Nerve stimulation (RNS) and exercise testing are the other electrodiagnostic examinations. Either high-frequency RNS (30-50 Hz) should be performed or a CMAP with a distal stimulation should be recorded to look for facilitation before and after 10 seconds of maximal voluntary exercise [24, 26]. Any increment greater than 40% is abnormal. It is calculated by;

$$increment = 100 \times \frac{Highest\ amplitude - Initial\ amplitude}{Initial\ amplitude} \quad (4.1)$$

Most patients with LEMS have increments greater than 100%. Slow RNS (3 Hz) can be performed on at least one proximal and one distal motor nerve. Decrements on slow

RNS are common in LEMS. However the evidence is not sufficient to different LEMS from MG [24].

Routine needle EMG of proximal and distal muscles especially being weak should be performed. Similar to MG, MUAPs may be unstable or short, small, and polyphasic with normal or early recruitment. SFEMG is usually not required in LEMS [36]. If applied, findings such as increased jitter and blocking will be consistent with a NMJ disorder. However, SFEMG cannot differentiate LEMS from other disorders of neuromuscular junction [24].

4.4.3 Botulism

Botulism is an acute neurologic disorder that causes potentially life-threatening neuroparalysis due to a neurotoxin produced by *Clostridium botulinum*. The toxin binds irreversibly to the presynaptic membranes of peripheral neuromuscular and autonomic nerve junctions. Toxin binding blocks acetylcholine release, resulting in weakness, flaccid paralysis, and, often, respiratory arrest [24, 26].

Routine motor and sensory nerve conduction studies should be performed at least two nerves, a motor and sensory nerve in one upper and one lower extremity. CMAP amplitudes are diffusely low with normal latencies and conduction velocities. Repetitive Nerve stimulation (RNS) and exercise testing are the other electrodiagnostic examinations. Either high-frequency RNS (30-50 Hz) should be performed or a CMAP with a distal stimulation should be recorded to look for facilitation before and after 10 seconds of maximal voluntary exercise [24, 26]. Any increment greater than 40% is abnormal. This increment value is computed by using (4.1). Most patients with Botulism have increments greater than 100%. Slow RNS (3 Hz) can be performed on at least one

proximal and one distal motor nerve as in myastenia gravis. Decrements on slow RNS might be seen in Botulism [24].

4.5 Myopathies

The myopathies comprise disorders in which primary pathology involves muscle tissue. Primary diseases of muscle can be genetic in origin such as muscular dystrophies and congenital myopathies, or can be acquired as in case of metabolic, endocrine and inflammatory myopathies.

4.5.1 Muscular Dystrophies (MD)

These are a group of inherited non-inflammatory muscle diseases having progressive clinical course without a central or peripheral abnormality. Muscular dystrophies are characterized by progressive skeletal muscle weakness, fiber degeneration, defects in muscle proteins, and the death of muscle cells and tissue [124, 130].

4.5.1.1 Duchenne Muscular Dystrophy

Duchenne Muscular Dystrophy is the most common childhood form of MD, as well as the most common of the muscular dystrophies overall, accounting for approximately 50 percent of all cases. It affects approximately one in 3,500 male births. Because inheritance is X-linked recessive (caused by a mutation on the X, or sex, chromosome), Duchenne MD primarily affects boys, although girls and women who carry the defective gene may show some symptoms [94]. About one-third of the cases reflect new mutations

and the rest run in families. Sisters of boys with Duchenne MD have a 50 percent chance of carrying the defective gene [26, 130].

Duchenne MD usually becomes apparent when an affected child begins to walk. Progressive weakness and muscle wasting (a decrease in muscle strength and size) caused by degenerating muscle fibers begins in the upper legs and pelvis before spreading into the upper arms. Other symptoms include loss of some reflexes, a waddling gait, frequent falls and clumsiness (especially when running), difficulty when rising from a sitting or lying position or when climbing stairs, changes to overall posture, impaired breathing, lung weakness, and cardiomyopathy (heart muscle weakness that interferes with pumping ability). Many children are unable to run or jump. The wasting muscles, in particular the calf muscle (and, less commonly, muscles in the buttocks, shoulders, and arms), may be enlarged by an accumulation of fat and connective tissue, causing them to look larger and healthier than they actually are and this is called pseudohypertrophy. As the disease progresses, the muscles in the diaphragm that assist in breathing and coughing may weaken [26, 130].

Patients may experience breathing difficulties, respiratory infections, and swallowing problems. Bone thinning and scoliosis (curving of the spine) are common. Some children are mildly mentally impaired. Between ages 3 and 6, children may show brief periods of physical improvement followed by progressive muscle degeneration. Children with Duchenne MD are typically wheelchair-bound by age 12 and usually die in their late teens or early twenties from progressive weakness of the heart muscle, respiratory complications, or infection [26, 130].

Duchenne MD results from an absence of the muscle protein dystrophin. And blood tests of children with Duchenne MD show an abnormally high level of creatine kinase, which is apparent from birth [125, 130, 134].

EMG demonstrates characteristic patterns of myopathy. Fibrillation potentials and positive sharp waves can be seen initially. As the disease progresses, fibrosis replaces muscle tissues. Low-amplitude, short-duration, complex MUAPs result from random loss of muscle fibers [24, 26, 130].

4.5.1.2 Becker Muscular Dystrophy

Becker MD is less severe than but closely related to Duchenne MD. Individuals with Becker MD have partial but insufficient function of the protein dystrophin. The disorder usually appears around age 11 but may occur as late as age 25, and patients generally live into middle age or later. The rate of progressive, symmetric on both sides of the body, muscle atrophy and weakness varies greatly among affected individuals. Many patients are able to walk until they are in their mid-thirties or later, while others are unable to walk past their teens. Some affected individuals never need to use a wheelchair. As in Duchenne MD, muscle weakness in Becker MD is typically noticed first in the upper arms and shoulders, upper legs, and pelvis [26, 130].

Early symptoms of Becker MD include walking on one's toes, frequent falls, and difficulty rising from the floor. Calf muscles may appear large and healthy as deteriorating muscle fibers are replaced by fat, and muscle activity may cause cramps in some people. Cardiac and mental impairments are not as severe as in Duchenne MD [26, 130].

Electromyography shows nearly symmetric abnormalities in the proximal muscles. Fibrillation potentials and complex repetitive discharges are abundant in paraspinal muscles. Small and polyphasic MUAPs show an early recruitment [26, 130].

4.5.1.3 Facioscapulohumeral Muscular Dystrophy (FSHD)

Facioscapulohumeral MD initially affects muscles of the face, shoulders, and upper arms with progressive weakness. Also known as Landouzy-Dejerine disease, this third most common form of MD is an autosomal dominant disorder. Life expectancy is normal, but some individuals become severely disabled. Disease progression is typically very slow, with intermittent spurts of rapid muscle deterioration. Onset is usually in the teenage years but may occur as late as age 40 [26, 130].

Muscles around the eyes and mouth are often affected first, followed by weakness around the lower shoulders and chest. A particular pattern of muscle wasting causes the shoulders to appear to be slanted and the shoulder blades to appear winged. Muscles in the lower extremities may also become weakened. Reflexes are impaired only at the biceps and triceps. Changes in facial appearance may include the development of a crooked smile, a pouting look, flattened facial features, or a mask-like appearance. Some patients cannot pucker their lips or whistle and may have difficulty swallowing, chewing, or speaking [26, 130].

Other symptoms may include hearing loss (particularly at high frequencies) and lordosis, an abnormal swayback curve in the spine. Contractures are rare. Some FSHD patients feel severe pain in the affected limb. Cardiac muscles are not affected, and the pelvic girdle is rarely significantly involved. An infant-onset form of FSHD can also cause retinal disease and some hearing loss [26, 130].

In initial stages, EMG demonstrates only limited abnormalities which may escape detection even in clinically weak muscles. In advanced cases, low-amplitude, short-duration polyphasic MUAPs with early recruitment may be observed [26, 130].

4.5.1.4 Limb-girdle Muscular Dystrophy

Limb-girdle MD refers to more than a dozen inherited conditions marked by progressive loss of muscle bulk and symmetrical weakening of voluntary muscles, primarily those in the shoulders and around the hips [124]. At least three forms of autosomal dominant limb-girdle MD (known as type 1) and eight forms of autosomal recessive limb-girdle MD (known as type 2) have been identified. Some autosomal recessive forms of the disorder are now known to be due to a deficiency of any of four dystrophin-glycoprotein complex proteins called the sarcoglycans [26, 124, 130].

The recessive limb-girdle muscular dystrophies occur more frequently than the dominant forms, usually begin in childhood or the teenage years, and show dramatically increased levels of serum creatine kinase. The dominant limb-girdle muscular dystrophies usually begin in adulthood. In general, the earlier the clinical signs appear, the more rapid the rate of disease progression. Limb-girdle MD affects both males and females. Some forms of the disease progress rapidly, resulting in serious muscle damage and loss of the ability to walk, while others advance very slowly over many years and cause minimal disability, allowing a normal life expectancy. In some cases, the disorder appears to halt temporarily, but symptoms then resume [26].

Weakness is typically noticed first around the hips before spreading to the shoulders, legs, and neck. Patients develop a waddling gait and have difficulty when rising from chairs, climbing stairs, or carrying heavy objects. Patients fall frequently and are unable to run. Contractures at the elbows and knees are rare but patients may develop contractures in the back muscles, which gives them the appearance of a rigid spine. Proximal reflexes (closest to the center of the body) are often impaired. Some patients also experience cardiomyopathy and respiratory complications. Intelligence remains

normal. Most persons with limb-girdle MD become severely disabled within 20 years of disease onset [26].

EMG shows typical myopathy findings such as low-amplitude and short-duration MUAPs in Limb-girdle myopathy [26, 130].

4.5.1.5 Emery-Dreifuss Muscular Dystrophy

Emery-Dreifuss MD primarily affects boys. The disorder has two forms: one is X-linked recessive and the other is autosomal dominant [26].

Onset of Emery-Dreifuss MD is usually apparent by age 10, but symptoms can appear as late as the mid-twenties. This disease causes slow but progressive wasting of the upper arm and lower leg muscles and symmetric weakness. Contractures in the spine, ankles, knees, elbows, and back of the neck usually precede significant muscle weakness, which is less severe than in Duchenne MD. Contractures may cause elbows to become locked in a flexed position. The entire spine may become rigid as the disease progresses. Other symptoms include shoulder deterioration, toe-walking, and mild facial weakness. Serum creatine kinase levels may be moderately elevated [26].

Nearly all Emery-Dreifuss MD patients have some form of heart problem by age 30, often requiring a pacemaker or other assistive device. Female carriers of the disorder often have cardiac complications without muscle weakness. Patients often die in mid-adulthood from progressive pulmonary or cardiac failure [26].

EMG shows typical myopathy findings such as low-amplitude and short-duration MUAPs in Limb-girdle myopathy [26, 130].

4.5.2 Congenital Myopathy

Congenital myopathy refers for any muscle disorder present at birth. These are conditions that have non-progressive or only slightly progressive muscular weakness. In the common, well-described congenital myopathies, mutations have been identified in genes that encode for muscle proteins. The loss or dysfunction of these proteins presumably leads to the specific morphological feature on muscle biopsy samples and to the clinical muscle disease [26].

These myopathies include nemaline myopathy, central core disease, centronuclear myopathy, minimulticore myopathy, congenital fiber-type disproportion, cytoplasmic body myopathy, fingerprint body myopathy, zebra body myopathy and congenital hypotonia with type I predominance [26, 130].

Electromyography (EMG) and nerve conduction studies (NCSs) should be performed in all patients in whom a congenital myopathy is suspected. In congenital myopathy, NCS findings are normal and EMG findings are either normal or show the typical small-amplitude, narrow-duration motor unit potentials (MUPs) that are seen in myopathies. Fibrillations and positive sharp waves are rare [26, 130].

4.5.3 Metabolic Myopathies

Metabolic myopathies refer to a group of hereditary muscle disorders caused by enzymatic defects due to defective gene insult [103]. Metabolic myopathies are heterogeneous conditions that have common abnormalities of muscle energy metabolism that result in skeletal muscle dysfunction. Most recognized metabolic myopathies are considered primary inborn errors of metabolism and are associated with known or postulated enzymatic defects that affect the ability of muscle fibers to maintain adequate adenosine triphosphate (ATP) concentrations. Traditionally, these diseases are grouped into glycogen storage disease, disorders of lipid metabolism, disorders of purine, or mitochondrial biochemistry [26, 130].

4.5.3.1 Acid Maltase Deficiency (Type II Glycogenosis)

This disorder is refered as Pompe's disease. It is an autosomal recessive disease where the deficiency leads to accumulation of glycogen in tissue lysosomes resulting in a vacuolar myopathy [26].

In the infantile type, severe hypotonia is developed just after the birth. These children die within the first year of the life due to the cardiac and respiratory failure [26].

EMG studies show insertional activity, fibrillation potentials, positive sharp waves and complex repetitive discharges. Polyphasic low-amplitude, short-duration MUAPs are recruited by mild voluntary contraction [26, 130].

4.5.3.2 Debrancher Deficiency (Type III Glycogenosis)

In this disorder being inherited as an autosomal recessive trait, breakdown of glycogen beyond the outer straight glucosyl chain is prevented by the absence of the debrancher enzyme. Thus, glycogen with short-branched outer chains, referred as phosphorilase-limit-dextrin accumulates in the liver. Hepatomegaly, episodes of hypoglycemia, elevated serum CK occur. Clinical features of myopathy may develop after hepatic symptoms are observed [26].

EMG may reveal profuse fibrillation potentials, complex repetitive discharges and small, short-duration MUAPs [26, 130].

4.5.3.3 Muscle Phosphorylase Deficiency (Type V Glycogenosis)

This disorder is called also McArdle's Disease and is an autosomal recessive condition. It results in a myophosphorilase deficiency that blocks the conversion of muscle glycogen to glucose during heavy exercise under ischemic conditions [135]. In infant, generalized hypotonia may lead to respiratory insufficiency and early death [26].

EMG studies may reveal fibrillation potentials and polyphasic MUAPs. Myotonic or complex repetitive discharges nay appear predominantly in paraspinal muscles [26, 130].

4.5.3.4 Muscle Phosphokinase Deficiency (Type VII Glycogenosis)

This disorder results from a defect in muscle phosphofructokinase preventing the conversion of fructose-6-phosphate to fructose-1-6-diphosphosphate [104]. In clinic, painful muscle contracture and myoglobulinuria occur. No abnormalities are revealed by EMG between attacks [26, 130].

4.5.3.5 Disorders of Lipid Metabolisms

Long-chain fatty acids are the major source of energy for the skeletal muscle during sustained exercise and fasting. The passage of these fatty acids through the mitochondrial membrane, for beta-oxidation, requires their binding with carnitine. Carnitine is synthesized mainly in the liver and actively transported into the muscle against a concentration gradient. Free fatty acids are first converted to acyl coenzyme A (CoA) compounds by the action of fatty acyl CoA synthetases. Then, the long-chain acyl CoA is bound to carnitine by acylcarnitine transferases, such as carnitine palmitoyltransferase I (CPT I). This occurs on the outer mitochondrial membrane [26, 130].

The new compound passes through the inner mitochondrial membrane by the action of acylcarnitine translocase. Within the mitochondrial matrix, carnitine palmitoyltransferase II (CPT II) splits the transferred compound to free fatty acids and carnitine. In the mitochondria, beta-oxidation of the long-chain fatty acids is then carried out. Carnitine deficiency, deficiency of carnitine palmitoyltransferases, or a defect in beta-oxidation of these fatty acids may lead to myopathies [26, 130].

In EMG studies, mild voluntary contractions that recruit small-amplitude, short-duration, polyphasic MUAPs is revealed. Fibrillation potentials and other form of spontaneous activity such as complex repetitive discharges are seen in the half of the patients [26, 130].

4.5.3.6 Mitochondrial Disease

Mitochondrial disorders encompass a group of disorders resulting from abnormalities of the respiratory chain. Needle EMG may show myotonic discharges in ADM, a relatively specific finding in patients with suspected metabolic myopathy. In general, needle EMG may reveal short-duration, low-amplitude motor unit action potentials [26].

4.5.4 Endocrine Myopathies

Major categories of endocrine myopathy include those associated with adrenal dysfunction (as in Cushing disease or steroid myopathy), thyroid dysfunction (as in myxedema coma or thyrotoxic myopathy), parathyroid dysfunction (as in multiple endocrine neoplasia), pituitary dysfunction, and islands of Langerhans dysfunction (as in diabetic myopathy from ischemic infarction of the femoral muscles). Steroid myopathy is the most common endocrine myopathy [26, 130].

In adrenal dysfunction, factors contributing muscle weakness in adrenal insufficiency include circulatory insufficiency, fluid and electrolyte imbalance, impaired carbohydrate metabolism, and starvation [26, 130].

In thyroid dysfunction, muscle weakness occurs most prominently in the adult forms of myxedema. Thyroid hormone excess also results in myopathy. Thyrotoxic myopathy is believed to be secondary to a disturbance in the function of the muscle fibers from increased mitochondrial respiration, accelerated protein degradation and lipid oxidation, and enhanced beta-adrenergic sensitivity due to excessive amounts of thyroid hormone [26, 130].

In parathyroid dysfunction, hypoparathyroidism causes tetany, with or without carpopedal spasm. The pathophysiology may involve either deficiency of parathyroid hormone or inability of the hormone to have an effect at end-receptors because of dysfunction of the hormone receptors. Hyperparathyroidism does not cause tetany but results in muscle wasting and myopathy (ie, proximal muscle weakness). The pathophysiology is oversecretion of hormone, frequently from a parathyroid adenoma. Myopathy related to parathyroid dysfunction appears to result from altered parathyroid hormone (PTH) level and impaired action of vitamin D [26].

In pituitary dysfunction, the myopathy from pituitary disease may be a result of secondary adrenal dysfunction and/or other endocrine disturbance such as thyroid dysfunction [26].

In thyroid myopathy, EMG studies shows increased insertional positive waves with some transient myotonic discharges. In parathyroid disease, EMG changes in tetany reveal the presence of MUAPs in doublets and triplets. In adrenal and pituitary disease, early recruitment of low-amplitude, short-duration MUAPs are demonstrated in EMG [24, 26, 130].

4.5.5 Myositis

Myositis is a general term for inflammation of the muscles. Many such conditions are considered likely to be caused by autoimmune conditions, rather than directly due to infection [26, 134]. The most common types of myositis are polymyositis (PM) and dermatomyositis (DM) [24, 26].

DM is a combination of skin rash and muscular weakness. Except the absence of skin lesions, the signs and symptoms of PM resemble to DM [26].

The electromyographic abnormalities in DM and PM are as follows: small-amplitude, short-duration polyphasic motor unit potentials, fibrillation potentials, positive sharp waves and insertional irritability, complex repetitive discharges. Electrical abnormalities are confined to paraspinal muscles [26, 130].

Another inflammatory disease of skeletal muscle is inclusion body myositis (IBM). Its distinctive characteristic is the pathologic feature that it possesses such as rimmed vacuole containing osmophillic membranous whorls and intranuclear and cytoplasmic inclusions. Clinically, IBM demonstrates slowly progressive weakness [24, 26]. Electromyographic abnormalities in IBM as in other myositis conditions comprise fibrillation potentials, positive sharp waves, complex repetitive discharges and low-amplitude, short-duration motor unit potentials with early recruitment [24, 130].

5. JUVENILE MYOCLONIC EPILEPSY

5.1 Introduction

Juvenile myoclonic epilepsy (JME) or impulsive petit ma1 was first described by Janz in 1957 [136]. Thus, juvenile myoclonic epilepsy (JME) is also known as Janz syndrome. Like childhood absence epilepsy (CAE), juvenile absence epilepsy (JAE), JME is a fairly common familial form of idiopathic generalized epilepsy, representing 5-10% of all epilepsies [15, 137-140]. Since they do not originate in one spot in the brain as does "focal" epilepsy but manifest over the whole brain simultaneously, these forms are referred as Idiopathic generalized epilepsies (IGE) [13, 141, 142].

5.2 Etiology

Several genes mutations have been suggested as predisposing factor toward a common idiopathic generalized epilepsy syndrome in human. CACNB4 encodes a calcium channel β subunit. β subunits are important regulators of calcium channel current amplitude, voltage dependence, and also regulate channel trafficking. Ala322Asp mutation in GABRA1 gene encoding the alpha1 subunit of the gamma-amino-butyric acid receptor subtype A ($GABA_A$) is another suggested genetic etiologic factor [143, 144].

JME has been shown to be linked to the HLA region on chromosome 6 [146, 147]. Linkage is a phenomenon observed in families and is defined by the cosegregation of two genes, in this case the HLA gene and the disease gene [12, 145]. The locus has been

designated EJM1 which predisposes to JME or related IGEs on chromosome 6p [13, 122, 138, 139].

5.3 Prevalence and Incidence

A prevalence of 0.5–1.0 per 1000 people in a population for juvenile myoclonic epilepsy has been reported with a female predominance [139, 148].

5.4 Symptoms

Juvenile myoclonic epilepsy is regarded as primary generalized epilepsy that consists of primarily generalized seizures [142]. The symptoms of JME consist of involuntary, bilateral, repeated, arrhythmic, irregular myoclonic jerks being predominantly in the arms and occurring shortly after awakening, tonic-clonic seizures and typical absence observed in one-third of the patients [11, 130, 141, 149]. Myoclonus may be revealed before other seizures occur [122]. Consciousness is usually not impaired, and sudden falls are unusual [136, 138, 150].

JME usually appear at the puberty where the mean onset is around 15 years of age. However, this may vary between 8 and 26 years [130, 141].

Photosensitivity which means that the myoclonic or tonic-clonic seizures are triggered by flickering or flashing light is common in people with JME. It can be defined as generation of polyspikes and slow-waves (Psw) in response to intermittent photic stimulation [136, 148]. Photosensitivity might occur in approximately 30% of cases, especially in women [140].

Provoking factors such as sleep deprivation, visual stimuli, emotional stress, alcohol and drug use, menstruation experienced in daily life increase the frequency of the jerks and often lead to generalized tonic clonic seizures (GTCS) [151].

5.5 Diagnosis

A full and accurate history is very important in diagnosing this type of epilepsy. The description of typical symptoms especially myoclonic jerks are informative. In addition to clinical findings and symptoms, scalp electroencephalography (EEG) will also be very helpful in making a diagnosis, as this type of epilepsy is associated with some specific EEG patterns [14], 42]. The EEG will usually show whether the person is also photosensitive [148].

The typical interictal EEG abnormality consists of a generalized 4- to 6-Hz spike or polyspike and slow-wave discharges 1 to 20 seconds in duration [138]. Usually, 1-3 spikes precede each slow wave. When absence seizures are also present, 3-Hz spike-and-wave activity may be seen in addition to the polyspike-and-wave pattern [137, 142].

The ictal EEG associated with myoclonic jerks typically reveals 10- to 16-Hz polyspike discharges. These may be preceded by spike and wave activity and are often followed by 1- to 3-Hz slow waves. The number of spikes is typically 5 to 20 and tends to be proportionately correlated with the clinical intensity of the seizure. These epileptic discharges may briefly persist, even after clinical activity has ceased [130, 151].

Absence seizures of JME may be associated with ictal EEG patterns consisting of 3-Hz spike-and-wave activity. Sometimes, these are preceded by 4- to 6-Hz polyspike-and-wave discharges, which slow to 3 Hz as the patient loses consciousness [151].

Hyperventilation, sleep deprivation and photic stimulation, often facilitate the appearance of epileptiform discharges [151]. Photic stimulation and video games which are the sources of intermittent flickering light stimultaion frequently precipitates spike-and-wave patterns that accompany generalized jerks [152]. This occurs in approximately 30% of patients with JME [148].

5.6 Treatment

The aim of pharmacotherapy is to minimize morbidity and prevent complications. The selection of antiepileptic drugs (AEDs) for the treatment of JME depends on patient's co-morbidities, preferences, prior history of adverse events, gender, etc. Primary AEDs are Lamotrigine, Topiramate, Levetiracetam and Zonisamide [130].

6. MATERIAL AND METHODS

6.1 Experimental Setup

Motor unit is the basic anatomic and functional unit of skeletal muscles [8]. Conventional electromyography is used in routine clinical studies to monitor motor unit action potentials. However, since the spike portion of the motor unit action potentials picked up by concentric needle electrodes is derived from 5–10% of all muscle fibers in a motor unit during this method, it cannot give an insight about the entire motor unit [8, 17]. Therefore, an electrophysiological method such as scanning EMG that is able to measure temporal and spatial properties of an entire mootor unit is required [79].

Scanning EMG has not been used in clinical examinations. On the other hand, it has been served as a research tool for better investigating the motor unit morphology and the generation of EMG signals in different parts of the motor unit. In this study, an experimental setup for scanning electromyography was designed and was established.

6.1.1 General Structure of the Experimental Setup

The experimental setup includes, a concentric needle EMG electrode, a single fiber EMG electrode, an actuator that provides the upward movement of the concentric needle EMG electrode, an EMG instrument, a digital-to-analog converter (DAC) installed into the EMG instrument, a data acquisition (DAQ) device and a notebook. The block diagram of the Scanning EMG setup that will be used in Scanning EMG method is illustrated in Figure 6.1.

6.1.2 Electrodes

This method is based on recording the electrical activity through the territory step by step via a concentric needle electrode (CNE) [8, 16, 17]. Two distinct electrodes are used in this experimental setup as a recording (scanning) electrode and a trigger electrode. The electrodes are CNE's with 37-mm length (Medelec ELITE Disposable, Vyasis Healtcare, Madison, USA).

The recording (scanning) electrode is moved upwards with a certain step size waiting and detects up a motor unit action potential (MUAP) signal at each step. The trigger electrode detects a single fiber action potential (SFAP) from a muscle fiber belonging to the motor unit under investigation. This SFAP is used to discriminate MUAPs fired by this motor unit being simultaneous with the muscle fiber generating SFAP as the trigger signal.

In previous studies performed with scanning EMG a single-fiber EMG (SFEMG) electrode was used as the trigger electrode [8, 16, 17]. However, since, it is disposable and much cheaper than SFEMG and requires lower contraction level compared to SFEMG electrode, the CNE electrode can be used for this purpose instead of the SFEMG electrode by simply adjusting the filter settings [60, 66].

These electrodes are connected to two distinct input channels of the preamplifier of the electromyography (EMG) instrument by means of needle holders that consist of an orientation-free hub with a shielded cable 106-cm long, terminated in a 5-pin 240° DIN plug VIASYS Teca, Madison, USA). The electrodes are connected to the needle holders via the hubs and, the needle holders to the EMG instrument through the DIN plug.

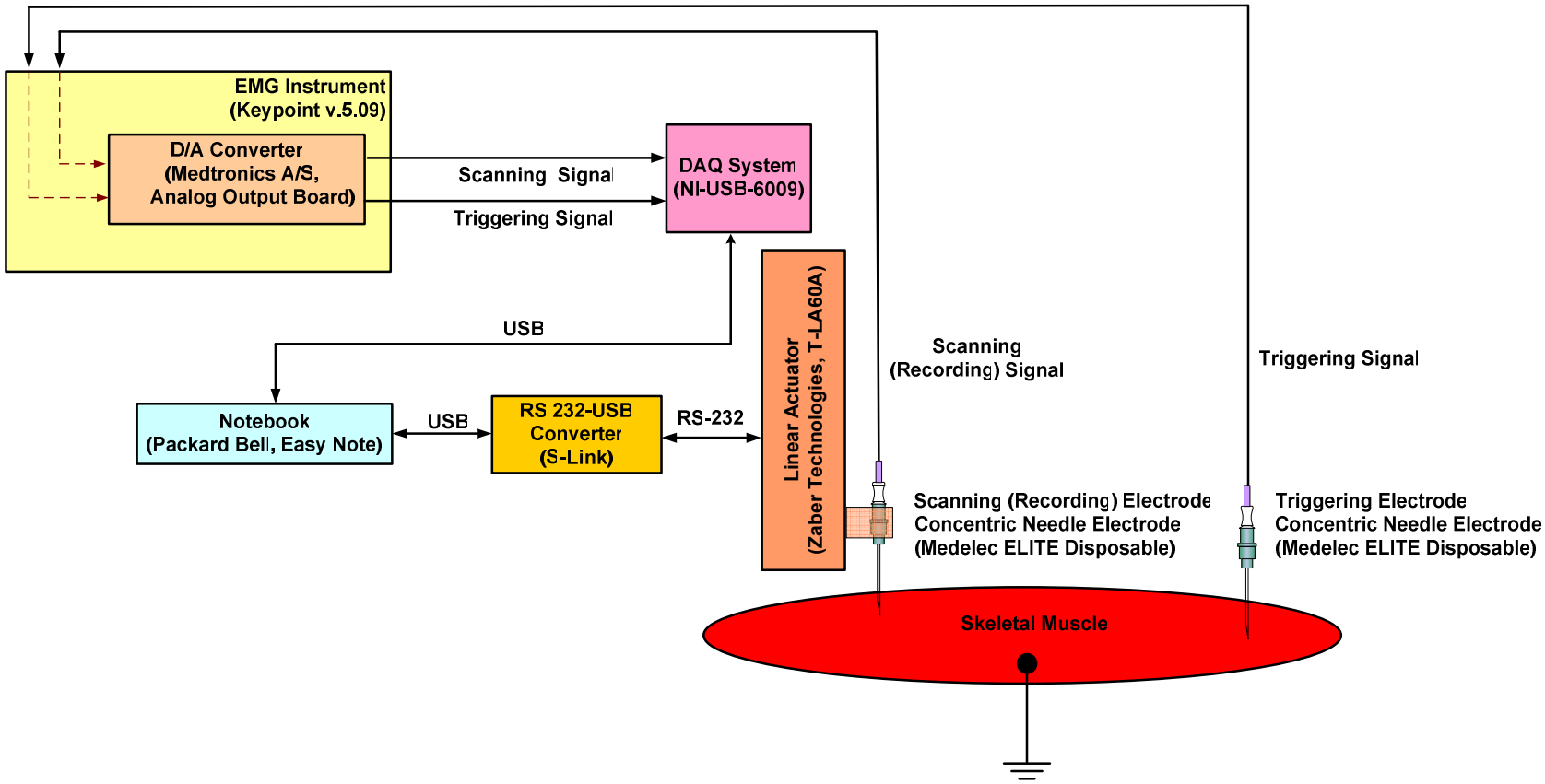


Figure 6.1 The block diagram of the Scanning EMG

6.1.3 Electromyography (EMG) Instrument

The MUAPs and the SFAPs picked up by both electrodes are in the order of mV. In addition, both electrodes act as antennas, and, receive also artifacts originating from sources of interference such as power lines.

EMG instrument (Keypoint version 5.09, Medtronics, Denmark) is used to amplify and to filter the acquired EMG signals by the record and trigger electrodes. Furthermore, these measured signals are displayed on the monitor of this instrument during either the insertion process of the needle electrodes into the muscle tissue or the recording process. This system has electrically isolated preamplifiers with CMRR more than 100 dB. The cut-off frequency for high-pass filter can be adjusted from 0.1 Hz to 3 kHz and that of low-pass filter may be set to 20 Hz. The balanced input is 200 MΩ. The amplitude resolution is 16 bits. The sampling rate is 48.0 kHz per amplifier. The system block diagram is drawn in Figure 6.2.

The electrodes coming from the patient are directly connected to the inputs of amplifier box. This circuit is isolated from other part of the EMG system. The amplification and filtering processes are achieved in this circuit.

The Dedicated Keyboard Assembly) not only comprises the keyboard functions but also the entire hardware for the auditory evoked potential (AEP) Stimulator, visual evoked potential (VEP) Stimulator, and Goggle Stimulator.

The PCI Front End Board is the interface between the Computer Unit Assembly (the PC) and the rest of the system. It has PC Card Controller circuit forming a bridge between the PCI Bus and the PC Card functions. The board is physically located in the Computer Unit Assembly where it is plugged into one of the PCI slots.

The PC platform of the Computer Unit Assembly is built around an active ATX Motherboard with Audio Controller, LAN Network Controller, and USB Ports on board. A chipset – Memory Controller and I/O Controller block on the System Block Diagram ties this and peripheral PC units together.

The filter settings for the channel of record electrode are adjusted to 10 Hz, and 10 kHz for high-pass and low-pass filter respectively. If SFEMG electrode were used as trigger electrode, the filter settings for trigger channel would have been adjusted to 500 Hz and 10 kHz for high-pass and low-pass filter respectively. Nonetheless, by adjusting the filter settings for this channel to 2 kHz and 10 kHz for high- and low-pass filter respectively, a concentric needle electrode can be used. Since they are cheaper than the SFEMG electrodes, require a lower contraction level compared to the SFEMG electrodes, are disposable and do not require sterilization after the use in each patient in contrast to SFEMG electrodes, CNEs are preferable [60]. SFEMG electrodes are re-usable and should be sterilized. The insulation material between the active electrode and the reference electrode can be damaged after the repetitive sterilization process by either autoclave or gas sterilization [26].

6.1.4 Analog Output Board

In order to be process and to record the signals coming from the electrodes by a computer, they have to be given as output from the EMG instrument. A D/A converter

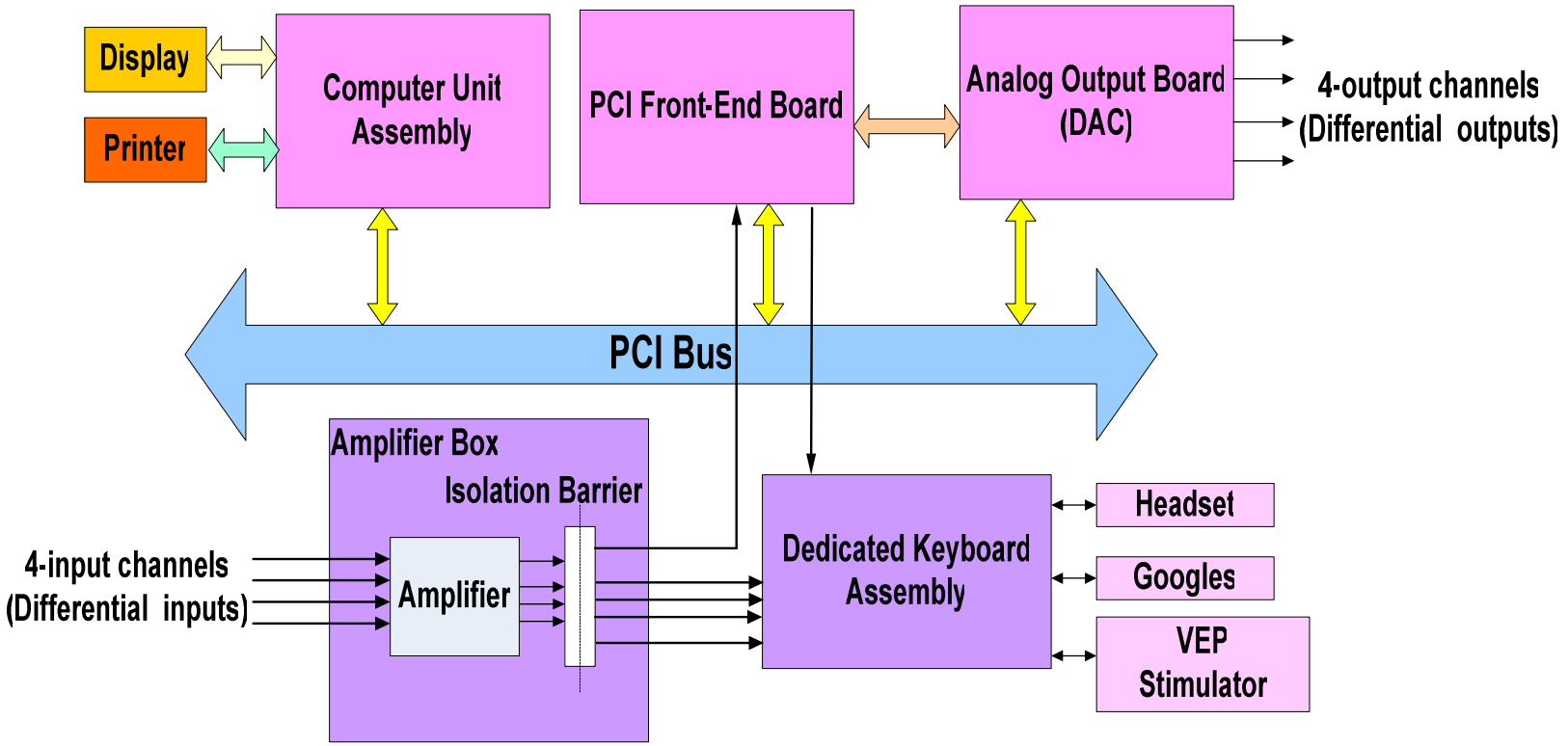


Figure 6.2 System block diagram of the Keypoint EMG system

(Medtronics A/S, Analog Output Board, Denmark) board with four output channels was installed into the EMG instrument. Two of these channels are used as outputs for either scanning signal or triggering signal.

This D/A converter board have a gain factor of 1000. The output voltage is +2.5 V. The output impedance is 1-kΩ. It has a 16-bit resolution. Sample rate is 48 kHz.

6.1.5 Data Acquisition (DAQ) System

In order to display and to store the data measured from the patients, the analog signals coming from the EMG instrument should be converted into the digital form. Thus, a data acquisition (DAQ) system is used for this purpose (NI-USB-6009, National Instruments, Austin, TX, USA). The DAQ system is shown in Figure 6.3.

Figure 6.3 Data Acquisition (DAQ) System.

The data signals are conveyed from the outputs of the D/A converter board to data acquisition (DAQ) system through two 50-Ω coaxial cables. These cables are connected to the outputs of the D/A converter board via the phono-sockets at one-end and to the DAQ system via a combicon jack at the other end. The analog inputs coming from EMG instrument are introduced to AI0+ and AI0- and AI1+ and AI1- signal ports corresponding to terminals 2, 3 and 5, 6 respectively. The connection of DAQ system to the computer system is achieved via a USB cable through the USB port of the computer.

This DAQ system has 14-bit resolution for differential mode. It has a maximum (aggregate) analog input sample rate of 48 kS/s (kilo samples per second) where the sampling rate is 24 kS/s for either scanning signal channel or triggering signal channel.

Input-ranges for differential mode are ±20, ±10, ±5, ±4, ±2.5, ±2, ±1.25, and ±1 V. The FIFO buffer size is 512 B. The timing resolution is 41.67 nanoseconds. The input impedance is 144-kΩ.

To utilize this DAQ device, the driver should be installed into the computer. It is supplied via two CDs shipped with the NI-USB 6009 DAQ system. Beside the driver, the libraries for high-level languages such as Visual Basic 6.0, Visual Basic.NET, Visual C etc. in NI-DAQxm software package. In order to configure and to test the NI-USB 6009 DAQ system, Measurement and Automation Explorer (MAX) is also installed.

6.1.6 Computer System and Interface Software

The upward and downward movements of the actuator are controlled by a notebook (Packard Bell, Easy Note, Wijchen, Netherlands). This is established by means of the

interface software created in Visual Basic 6.0 where parameters such as step size, step count, step period and the relative position of the electrode can be adjusted. This software is used also to control the DAQ system during the recording process by adjusting parameters such as number of samples, frequency (or sampling rate), voltage range of the acquired signal. This interface software that consists of Setting Menu, File Menu, Manual Menu, Auto Menu and Position Display Menu and is shown in Figure 6.3.

In Setting Menu, the communication port through which the communication between the actuator and the computer (Packard Bell Easy Note) is chosen. The chosen port is activated by pushing "Open Com Port" button. The movement unit can be chosen as either in steps or in microns. The value of home position can be entered into text box in microns. This feature is added to protect the mechanical systems establishing the upward and downward movement of lead screw. In normal case, it retracts until it triggers its

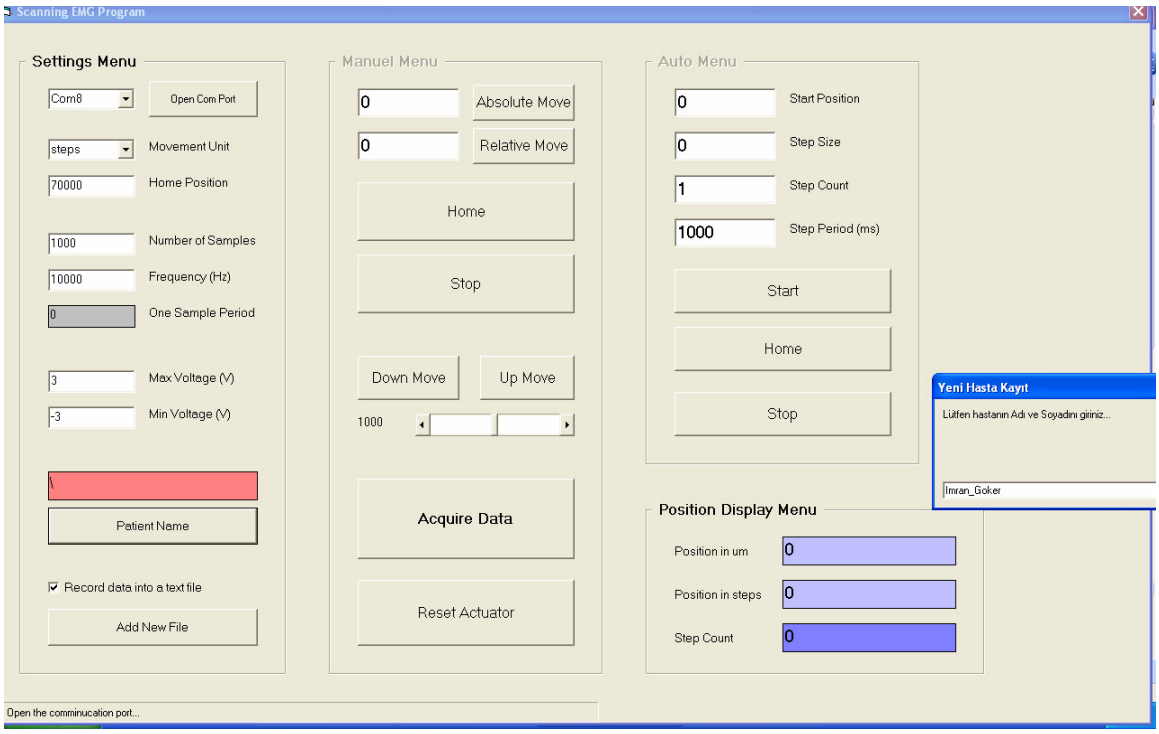


Figure 6.4 The console of the interface software.

internal home switch. There will be no problem if no needle attachment is attached to the lead screw of the actuator.

When this needle attachment is attached to the lead screw, if there were no home-position distance, it could damage the mechanical system located inside the housing of the linear actuator while forcing the lead screw to return zero position. The home position value is set to 70000 microns as a default value that is sufficient to insert the needle attachment.

The sampling rate can be determined by the value entered into text box with the label “Number of samples”. The sampling time changes automatically with the change in value of “Number of samples”.

The voltage that will be measured can be adjusted by entering values to max voltage and min voltage text boxes.

The File Menu is the menu where a folder is opened for each patient being studied. The name of the patient from whom data is acquired is entered in this menu. A folder is created in root directory (C:\...). When “Add New Patient” button is pushed, an input box is opened that asks the user to enter the name and the surname of the patient. Afterwards, a folder labeled with the patient name and surname is created in the “emgdata” directory located in the root directory of the notebook. Then by pushing “New File” button, the “Acquire Data” button is activated. The acquisition is established through the DAQ system during the automatic movement of the linear actuator. These data are stored in the file created by pushing “New File” button in the C:\emgdata directory. This folder is labeled with the format yymmdd_hhmmss.csv yymmdd indicates the date information (y: year; m: month, d: day) of the acquired data and hhmmss indicates the time information of that data (h: hour, m: minute, s: second).

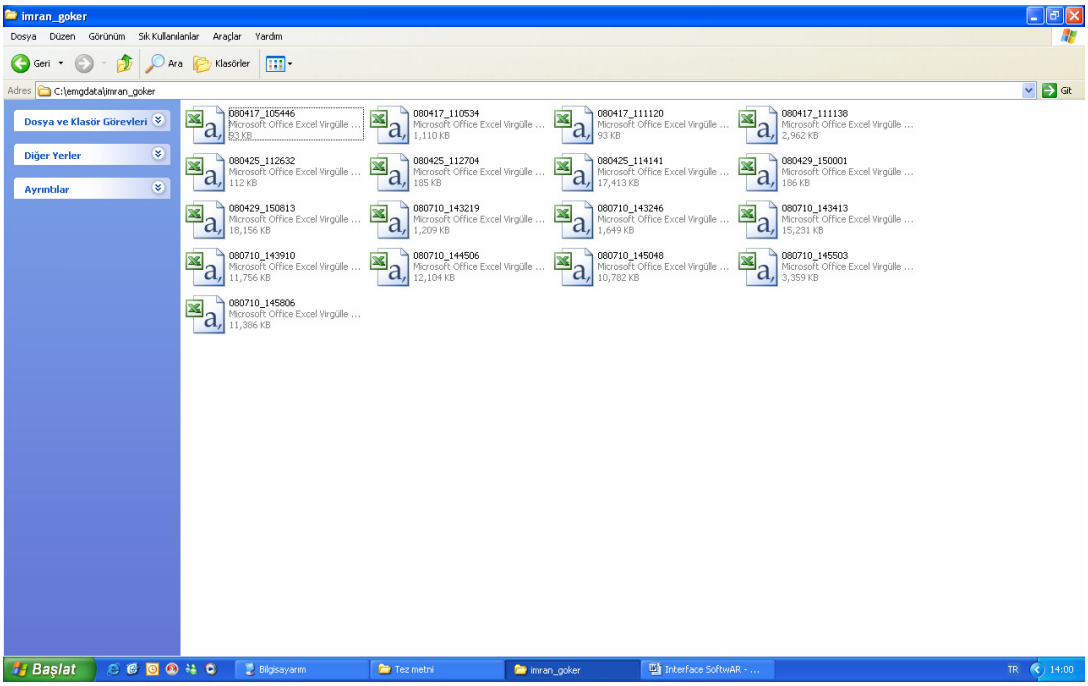


Figure 6.5 Data in .csv format stored in the root directory of the computer.

The Manual Menu is used to move the actuator to a desired position. This is achieved by entering a value for position and then by pushing absolute move button. Thus, the actuator moves to the absolute position from the home position. In order to move the actuator from the last position to another position without returning back to home position, the relative move button is used. This is established by entering relative position value to text box which is adjacent to the relative move button and then by pushing this button. The actuator can be moved back by pushing home button. In case of emergency, the movement of the actuator can be ceased by pushing stop button. The continuous movement of the actuator either in downward or upward direction is performed by pushing Downward Continuous Move button or Upward Continuous Move button. The actuator moves while one of these buttons is pushed and it stops when they are not pushed.

The Auto Menu enables the user to set the incremental movement of the actuator. A position value is entered to the text box being adjacent to Start Position label. This value determines from where the actuator will start the incremental movement. The step size value entered to the text box adjacent to Step Size label determines the distance that will be traveled in each step. The step count value determines how many steps will be taken during the incremental movement Step period value determines the time that will be spent in each step in milliseconds. After entering all these values, the incremental movement is activated by pushing Start button. The actuator moves continuously to the start position and then moves incrementally until last step count. When the last is finished, the actuator moves to the home position. Before initiating the incremental movement of the actuator, Acquire Data button is should be pushed to initiate the data acquisition process during the incremental movement.

In case of emergency, the incremental movement can be ceased by pushing Stop button in Auto Menu. After stopping the movement, the actuator can return home position by pushing Home button.

The position of the actuator in μm, the position of the actuator in steps and the step counts are displayed in the Position Display Menu.

While carrying the actuator, its lead screw should be kept in zero position. Therefore, the needle attachment should be removed from the lead screw. Then, in order to protect the lead screw from radial loads, it should be pulled to zero position. This is achieved by pushing Reset Actuator button.

The computer system is also used to store the text file of the acquired data in .csv format. These data are then used to reconstruct the three-dimensional electrophysiological cross-sectional map of the motor units under investigation.

6.1.7 Linear Actuator

The upward movement of the scanning electrode is achieved via a linear actuator (Zaber Technologies, T-LA60A, British Columbia, Canada). This actuator has a motion range of 60 mm and a resolution of 0.1 μm Since the step size of the upward movement of the scanning electrode was 50 μm and multiples thereof this resolution value satisfies the step size requirement of the study. In this study, in order to ensure that the total distance traversed by the CNE in the muscle is 2.5 to 3 cm, the step count is set to 250 to 300 steps and the step size is chosen as 100 μm at the console of the interface software. The linear actuator is connected to the notebook via a RS-232 cable. However, since the notebook has no RS-232 port, a RS-232-to-USB converter (S-Link, China) is used to adapt the connection of the linear actuator to the notebook. This linear actuator is shown in Figure 6.5

6.1.8 Needle Attachment

The movement of the concentric needle electrode that will be used to acquire data from the subjects is achieved by means of the upward movement of the lead screw of the linear actuator. A needle attachment is required to affix the concentric needle electrode to the needle electrode.

Figure 6.6 Linear actuator used in the experimental setup of Scanning EMG.

The portion of this needle attachment where the concentric needle electrode is inserted is manufactured from cast-amide. This portion is carved in a fashion that the plastic part of the concentric needle electrode fits. The strength of this cast-amide portion is ensured by a metal frame.

A hole is opened vertically through the needle attachment in order to the lead screw of the linear actuator. The lead screw is tightened by means of a screw to fix the needle attachment. The needle attachment is represented in Figure 6.6 and is shown in Figure 6.7.

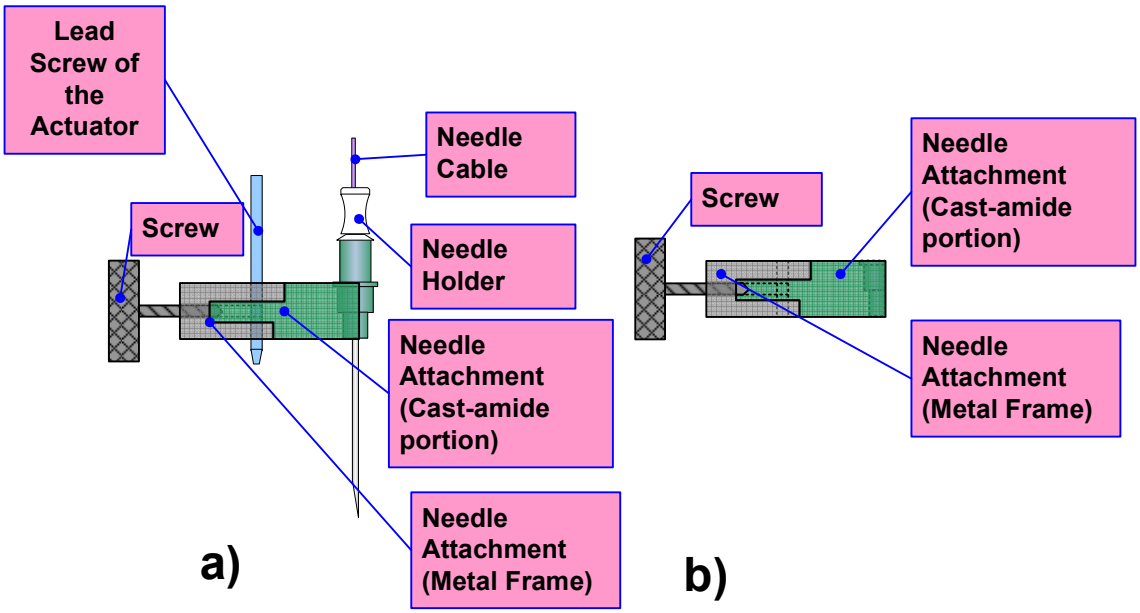


Figure 6.7 a) Needle attachment with the concentric needle electrode **b)** Needle attachment without the concentric needle electrode.

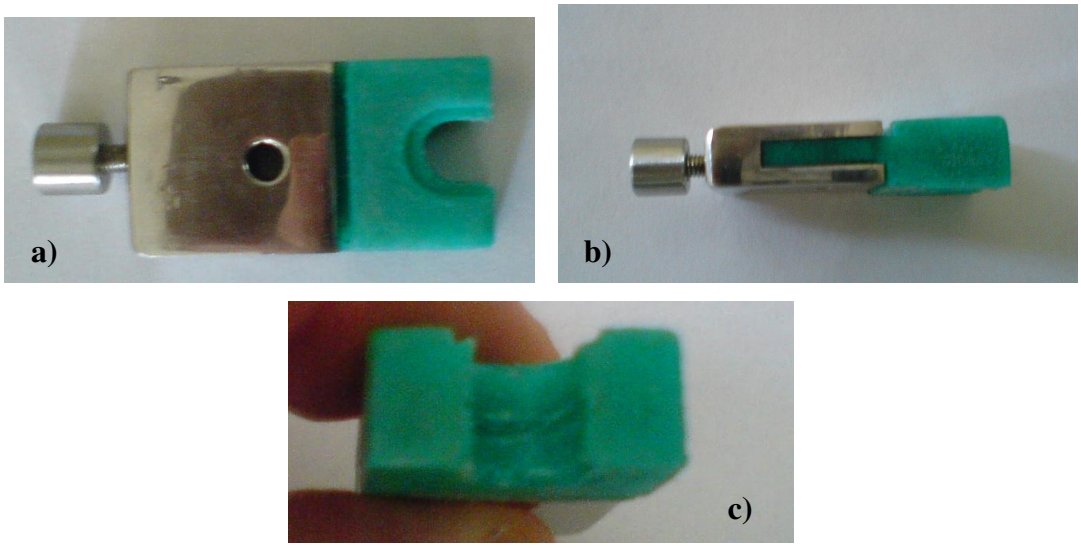


Figure 6.8 a) Needle attachment (top view) **b)** Needle attachment (size view) **c)** Needle attachment (front view).

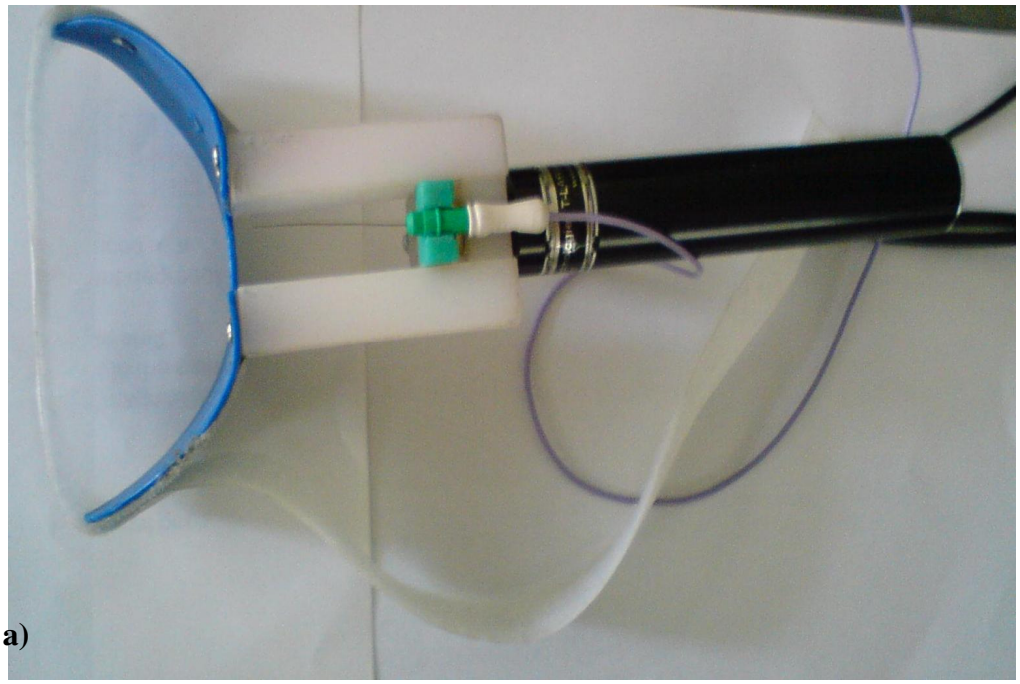
6.1.9 Support Attachment

In order to be able to hold the actuator during the movement of its lead screw and hence the movement of the concentric needle electrode, a simple support attachment was designed and is manufactured from Teflon. It is designed in a fashion that it permits the vertical motion of the needle attachment. It is attached to the housing of the linear actuator by tightening it via a hexagonal nut. It has two curved pieces to grasp the arm and a strap to tighten the attachment. It is used to affix the linear actuator to the arm of the subject. The support attachment is shown in Figure 6.8 and illustrated schematically in Figure 6.9.

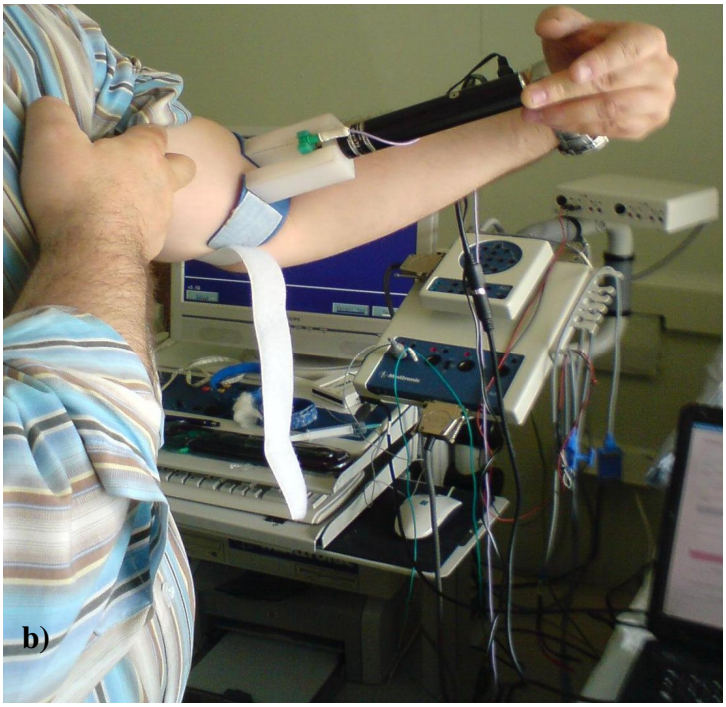
6.2 Three-Dimensional Plots of Motor Unit Territory

The aim of this experimental setup is to acquire data that will be processed to plot the three-dimensional electrophysiological maps of the motor units under the investigation. Both trigger and scanning data are stored in the root directory of the computer in .csv format.

Scanning data acquired during 200-milisecond sweeps in each steps of the CNE moved upward by the linear actuator are used to reconstruct the waveform of the MUAPs generated by the motor unit in question. However, the scanning data may consist also MUAPs that are not time-locked with the single-fiber action potential (SFAP) produced by the motor unit under investigation and that come from other motor units.



a)



b)

Figure 6.9 a) The actuator with support attachment, needle attachment, and the concentric needle electrode **b)** The actuator with support attachment, needle attachment, and the concentric needle electrode connected to the experimental setup and to EMG system to acquire data from biceps brachialis muscle

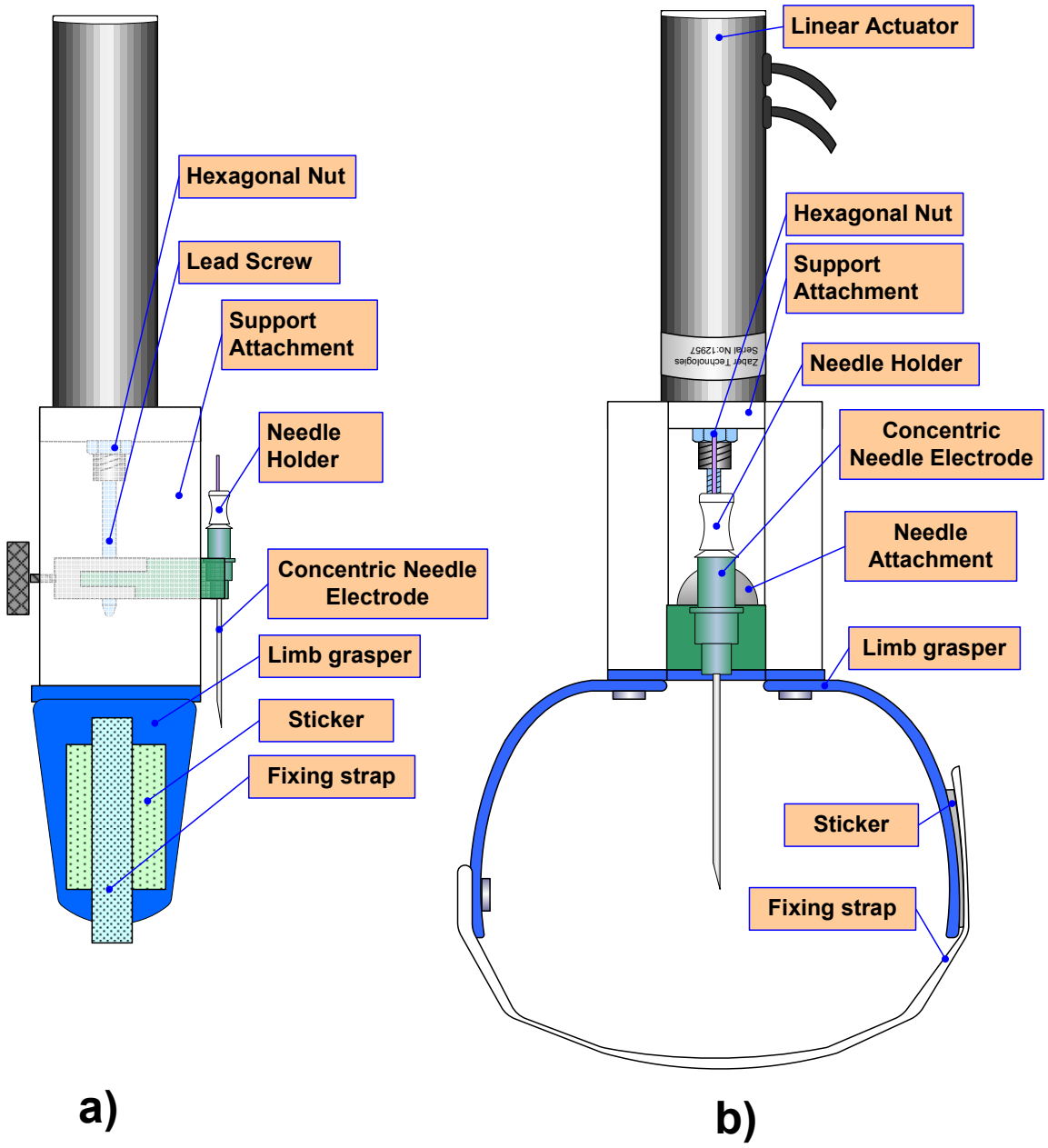


Figure 6.10 a) The actuator with support attachment, needle attachment, and the concentric needle electrode (side view) **b)** The actuator with support attachment, needle attachment, and the concentric needle electrode (front view).

Figure 6.11 Experimental Setup of Scanning EMG connected to EMG instrument

An M-File in MATLAB version 7.2 is created to extract the scanning data synchronous with the SFAPs generated by the muscle fiber of the motor unit of interest. Trigger data represent these SFAPs.

The spurious activity on individual sweeps is rejected by means of a median filtering process described in earlier studies [16, 17]. MATLAB version 7.2 possesses median filtering property in its library.

As a result, three-dimensional electrophysiological cross-section of motor units of interest is plotted. The lengths of cross-section of these motor units are calculated by counting the steps where electrical activities are present and then by multiplying by 100-μm step size.

This M-File also finds the maximum amplitude of each motor unit. Both lengths of cross-sections and maximum amplitudes are used as statistical variables to compare the juvenile myoclonic epilepsy (JME), normal control (NC) and spinal muscular atrophy (SMA) groups. The flow chart of the algorithm of this M-File is represented in Figure 6.12.

Three-dimensional electrophysiological cross-sections of MU territories of a patient with JME, of a healthy volunteer, and that of a patient with SMA are illustrated in Figure 6.13, Figure 6.14, and Figure 6.15 respectively.

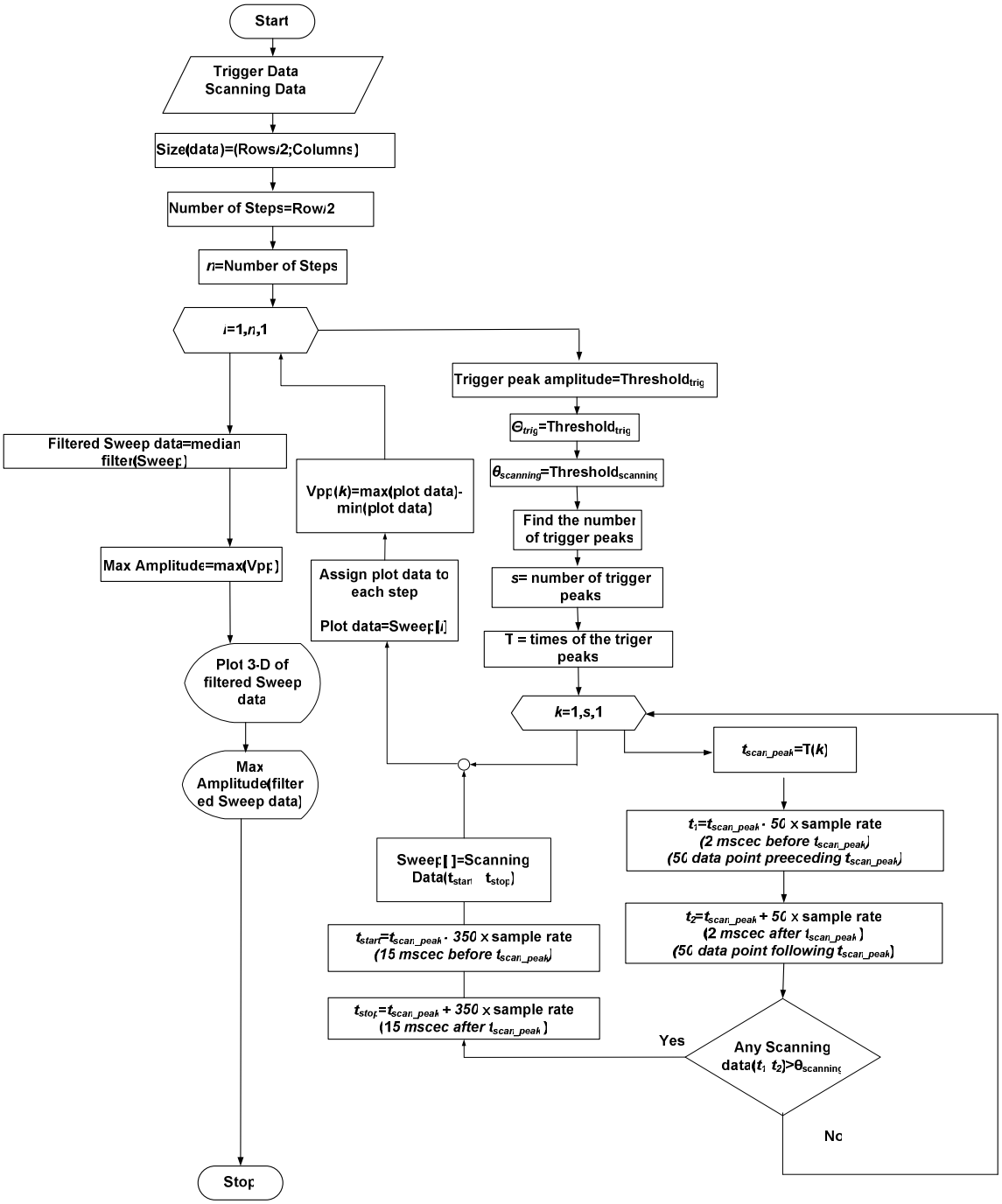


Figure 6.12 Flow chart of the algorithm of the M-File software created in MATLAB 7.2

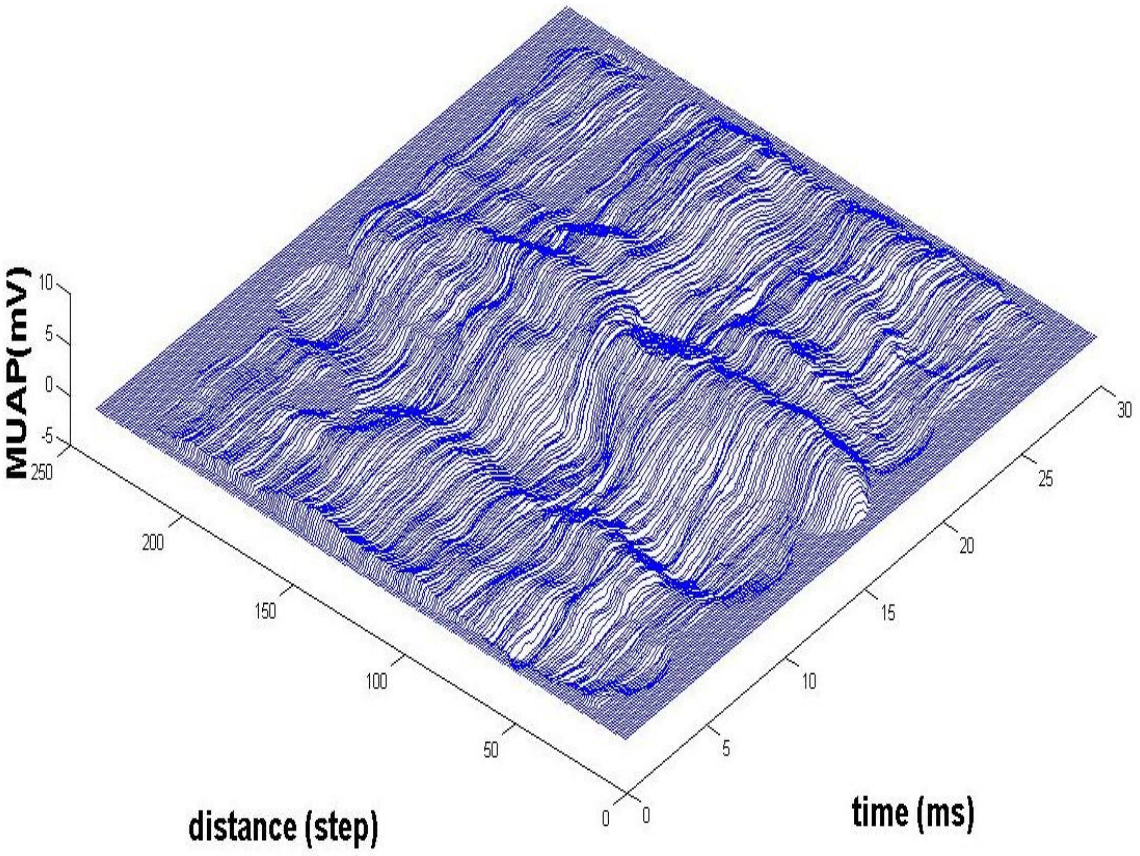


Figure 6.13 Three-dimensional electrophysiological cross-sections of MU territories of a patient with JME.

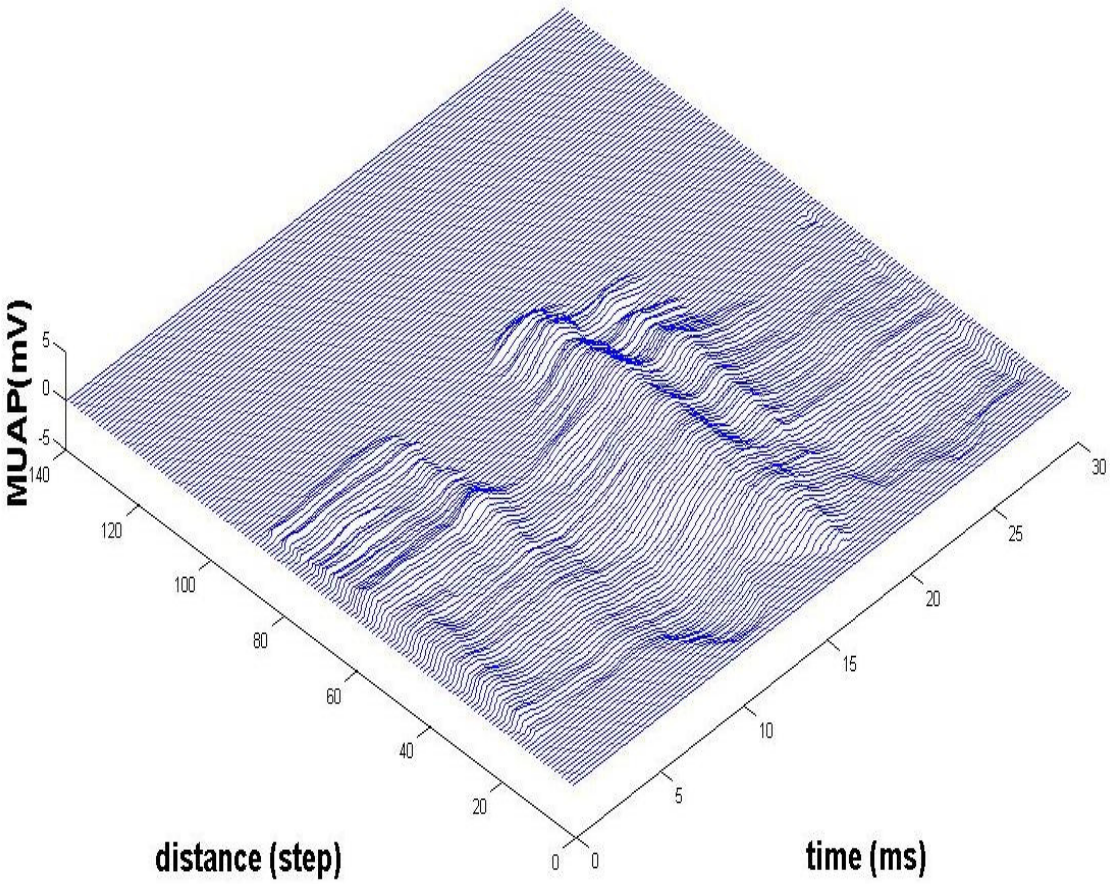


Figure 6.14 Three-dimensional electrophysiological cross-sections of MU territories of a healthy volunteer.

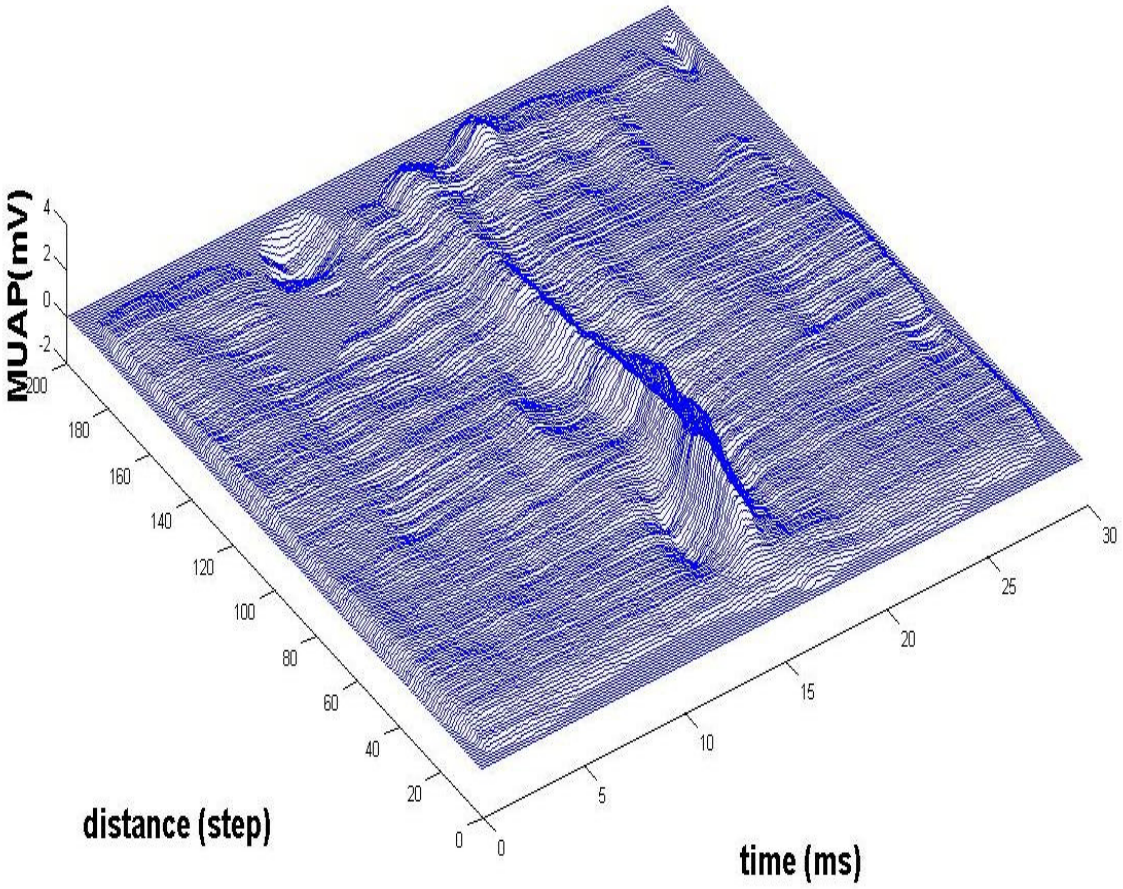


Figure 6.15 Three-dimensional electrophysiological cross-sections of MU territories of a patient with SMA.

6.3 Clinical Measurements

Three subject groups that consist of patients with JME, healthy subjects as normal control as NC group and patients with spinal muscular atrophy (SMA) were studied. JME, NC and SMA groups comprised nine, ten and three individuals that fulfill the inclusion criteria respectively. All individuals enrolled to this study were requested to read and to sign an informed consent.

The measurements that were performed with scanning EMG were established at the EMG laboratory of the Neurology Department at Istanbul Medical Faculty of İstanbul University. Scanning EMG recordings were performed on the biceps brachii muscle of the subjects lying in supine position as shown in Figure 6.16. The ground electrode of the EMG instrument was attached to the wrist of the subject.

First, the needle attachment was affixed to the lead screw of the linear actuator. The number of digitization samples in a single step was selected as 4700 samples on the console of the interface software. The sampling frequency was selected as 23.5 kHz on the console of the interface software. The sweep period was automatically calculated to 200 milliseconds. The period of a single step where the actuator waits before pulling the scanning electrode for the next step was set to 500 milliseconds. The step size and the number of steps were set to 100 μm and 200 to 300 steps respectively. The CNE used as scanning electrode was inserted into this needle attachment and was connected to the EMG instrument via the needle holder. The triggering electrode was inserted into the biceps brachialis (BB) muscle until a single fiber action potential (SFAP) of a muscle fiber belonging to any MU was detected, then, the electrode was left fixed. Afterwards, the linear actuator was fixed to the arm of the subjects by means of the support attachment in the fashion that the scanning electrode will be inserted into muscle as close as possible to the triggering electrode in order to pick up the MUAPs from the same MU

as that of the SFAP picked up by the triggering EMG. This distance was approximately 5 mm. horizontally. The scanning EMG was inserted 1cm deeper than the trigger electrode. Both triggering and the scanning signals were monitored in two channels on the display of the EMG instrument in real time to achieve the correct location of both electrodes. Once the electrical activity synchronous with that of triggering electrode was found, the scanning electrode was pushed away inside the MU until a position was reached where no further spike components of the activity of the scanning signal were detected. This was the lower boundary of the MU territory. Positioning procedure of both electrodes usually takes about 1 minute. Then, the scanning electrode was pulled step by step by the linear actuator during a slight voluntary contraction until no further spike component was seen on the display of the EMG instrument in the channel of the scanning signal. This step represented the upper boundary of the MU territory. Then, the scanning electrode was pulled to complete the measurement procedure and to repeat the insertion procedure described as above in order to measure the electric activity of another MU. 5 to 10 distinct measurements were performed for each subject. Each measurement lasted 2 minutes.

6.4 Statistical Evaluation

When the number of motor units is considered, the descriptive statistics of the variables such as length of cross-section of the motor units, maximum amplitude of the motor units, and age of the subjects are expressed as mean ± S.D.

Results from the patşents were compared in terms of number of motor units in pairs such as JME and NC, JME and NC, and SMA and NC using a parametric test such as Student's t-test with a confidence of interval of 95% in order to investigate their level of significance.

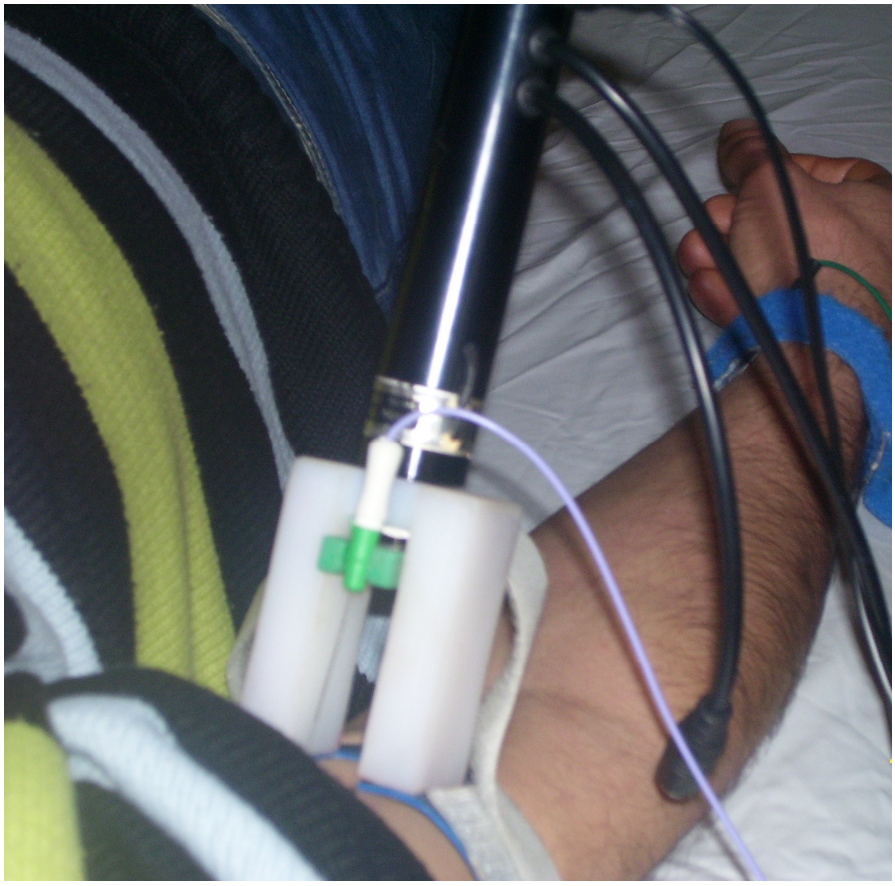


Figure 6.16 Recording process of EMG signal from the biceps brachialis muscle of a subject lying in supine position.

Since there are three groups with two independent variables each, length of cross-section of motor unit and the maximum amplitude of the motor unit, one-way ANOVA test and a post hoc test such Tukey's HSD test were applied.

In addition, the correlation between the length of cross-section and the maximum amplitude was also investigated by computing the coefficient of determination.

The length of cross-section of the motor unit and the maximum amplitude of motor unit were also studied on the basis individual subjects. The mean and the median of both length of cross-section of motor units and the maximum amplitude of the motor units were computed for each subjects. The values are expressed as mean ± S.D.

All groups were compared in terms of subjects included into the study in pairs such as JME and NC, JME and NC, and SMA and NC using a parametric test such as Student's t-test with a confidence of interval of 95% in order to investigate their level of significance. On the other hand, since the sample sizes are small for each group in terms of the subject number, these groups were studied also by using Mann-Whitney U-Test.

These three groups were compared by means of Kruskal-Wallis one-way analysis of variance as a non-parametric test.

7. RESULTS

The descriptive statistics were computed both in terms of number of motor units that were studies and in terms of the patients that were included into the study. These values were classified as JME group, NC group and SMA group in both cases.

In JME group, 52 motor units were studied. The length of cross-section ranges between 0.6 and 2.49 cm. with the mean value 1.4665 ± 0.51 cm. Maximum amplitude ranges from 1.94 to 18.40 mV. The mean value is 7.6843 ± 3.17 mV.

When NC group is considered, length of cross-section lies between 0.36 and 1.85 cm with mean value 1.1149 ± 0.375 cm. Maximum amplitude ranging between 1.28 and 12.20 mV has mean value 4.6594 ± 2.54 mV.

In SMA group, length of cross-section lying between 1.08 and 2.00 cm has mean value 1.7167 ± 0.2434 cm. Maximum amplitude ranges between 1.83 and 47.55 mV and has mean value 10.1396 ± 11.2646 mV.

These descriptive statistics that are computed for the number of motor units are tabulated in Table 7.1.

Table 7.1

The descriptive statistics computed in terms of number of motor units

	JME group (*n=52*)		**NC group (*n=51*)**		**SMA group (*n=15*)**	
	LCS (cm)	Max. Amplitude (mv)	LCS (cm)	Max. Amplitude (mv)	LCS (cm)	Max. Amplitude (mv)
Min.	0.6	1.94	0.36	1.28	1.08	1.83
Max.	2.49	18.40	1.85	12.20	2.00	47.55
Mean±S.D.	1.4665±0.5	7.6843±3.17	1.1149±0.375	4.6594±2.54	1.7167±0.2434	10.1396±11.2646

LCS= Length of cross-section

In JME group, nine patients were included into the study. Therefore, nine values were computed. The minimum and the maximum of the means of the lengths of cross-section of motor units are 0.78 and 2.03 cm respectively. The minimum and maximum values of the medians of lengths of cross-section of motor units are 0.74 and 2.04 cm respectively. The mean of the means of the lengths of cross-section of motor units is 1.4538 ± 0.44 cm. The mean of the medians of lengths of cross-section of motor units is 1.4378 ± 0.44 cm. The minimum and the maximum of the means of the maximum amplitudes of motor units are 4.82 mV and 12.30 mV respectively. These values for the medians of the maximum amplitudes are 4.32 and 11.32 respectively. The mean of the means of the maximum amplitudes of motor units is 7.56 ± 2.48 mV. The mean of the medians of the maximum amplitudes of motor units is 7.05 ± 2.29 mV.

In NC group, ten patients were included into the study. Thus, ten values for descriptive statistics were computed. The minimum and the maximum of the means of the lengths of cross-section of motor units are 0.71 and 1.50 cm respectively. The minimum and maximum values of the medians of lengths of cross-section of motor units are 0.70 and 1.62 cm respectively. The mean of the means of the lengths of cross-section of motor units is 1.1459 ± 0.23 cm. The mean of the medians of lengths of cross-section of motor units is 1.1455 ± 0.28 cm. The minimum and the maximum of the means of the

maximum amplitudes of motor units are 1.76 mV and 7.30 mV respectively. These values for the medians of the maximum amplitudes are 1.68 and 7.68 mV respectively. The mean of the means of the maximum amplitudes of motor units is 4.4266 ± 2.1 mV. The mean of the medians of the maximum amplitudes of motor units is 4.4585 ± 2.26 mV.

In SMA group, three subjects are included. In this context, three values were found to be used in the computation of the descriptive statistics. The minimum and the maximum of the means of the lengths of cross-section of motor units are 1.56 and 1.89 cm respectively. The minimum and maximum values of the medians of lengths of cross-section of motor units are 1.53 and 1.90 cm respectively. The mean of the means of the lengths of cross-section of motor units is 1.7167 ± 0.16 cm. The mean of the medians of lengths of cross-section of motor units is 1.7067 ± 0.18 cm. The minimum and the maximum of the means of the maximum amplitudes of motor units are 2.89 mV and 16.96 mV respectively. The values for the medians of the maximum amplitudes are 2.90 and 10.81 mV respectively. The mean of the means of the maximum amplitudes of motor units is 10.139 ± 7.04 mV. The mean of the medians of the maximum amplitudes of motor units is 7.8905 ± 4.34 mV.

These descriptive statistics that are computed for the number of subjects are summarized in Table 7.2.

Table 7.2

The descriptive statistics computed in terms of number of subjects

	JME group (n=9)			NC group (n=10)			SMA group (n=3)		
	Min.	Max.	Mean±S.D.	Min.	Max.	Mean±S.D.	Min.	Max.	Mean±S.D.
Mean LCS (cm)	0.78	2.03	1.4538±0.44	0.71	1.50	1.1459±0.23	1.56	1.89	1.7167±0.16
Median LCS (cm)	0.74	2.04	1.4378±0.44	0.70	1.62	1.1455±0.28	1.53	1.90	1.7067±0.18
Mean max. amplitude (mV)	4.82	12.30	7.56±2.48	1.76	7.30	4.4266±2.1	2.89	16.96	10.139±7.04
Median max. amplitude (mV)	4.32	11.32	7.05±2.29	1.68	7.68	4.4585±2.26	2.90	10.81	7.8905±4.34

LCS= Length of cross-section

8. DISCUSSION

Since JME and a locus regulating the spinal development are both mapped to chromosome 6, it is suggested previously that an anterior horn cell disorder may accompany JME [12-14]. Because such a disorder will affect a motor neuron that results in alterations in the anatomical and functional structure of the motor unit, these alterations are expected to be found by investigating the electrical activity of the motor unit. Also, a subclinical anterior horn cell involvement has been revealed in a recent study by means of conventional EMG and quantitative electrophysiological techniques such as IPA and TAA [14].

Because an increase in MU size is expected due to the reinnervation of this MU by the collateral sprouts from the adjacent motor neurons in case of anterior horn cell involvement, a sensitive method reflecting the electrical activity of the entire MU such as a Macro EMG was used in a previous study [15]. On the other hand, motor unit may be "large" in two respects: either with higher fiber density but with constant territory, or occupying a large territory but with constant fiber density [153]. Since, it gives quantitative aspects of the size and it would have differentiated between reinnervation and other reasons for the enlargement by means of its inbuilt single-fiber EMG (SFEMG) recording [2, 15, 26, 72]. It is possible to assess also the fiber density (FD) of a motor unit beside the macro MUAP [15]. Previously, the fiber density of the MU was found almost normal in JME cases although the macro MUAPs were increased. This suggests that this evidence is the due to the large motor units with normal organization of individual muscle fibers rather than reinnervation [15].

Also, a decrease in the number of motor axons to the muscle under the investigation implies the preponderance of the large MUs and therefore a motor unit number estimate (MUNE) analysis was performed previously in JME patients which concluded that

MUNEs of JME patients were nearly half of those of the normal subjects [15]. However, although both techniques may help to estimate the MU size neither macro-EMG nor MUNE techniques do not give an insight for the MU territory [15].

Another electrophysiological technique that might reflect the temporal and the spatial characteristic of the motor unit territory can be used for this purpose. Scanning EMG gives the electrophysiological cross-section of the motor unit under the investigation.

In addition to the parameters used with conventional EMG such as amplitude and duration, new parameters are introduced with this technique. These are length of MU cross-section, fractions of MUs, silent periods, polyphasic and complex portions of MUAPs, maximum duration and the maximum amplitude [8]. In this study, maximum amplitude and length of cross-section were used in order to compare JME, NC and SMA groups. Since there is increase in the length of cross-section of the motor in patients with SMA due to the reinnervation, a subject group with SMA was also included as a control group in this study.

As shown in Figure 8.1, although the SMA group seems to be positively skewed, three groups are almost symmetrically distributed. JME there was a highly significant difference between these groups in terms of length of cross-section.

All of these groups are symmetrical in terms of maximum amplitudes as illustrated in Figure 8.2. The median values of SMA and JME groups are similar. However, they are significantly different from that of NC group.

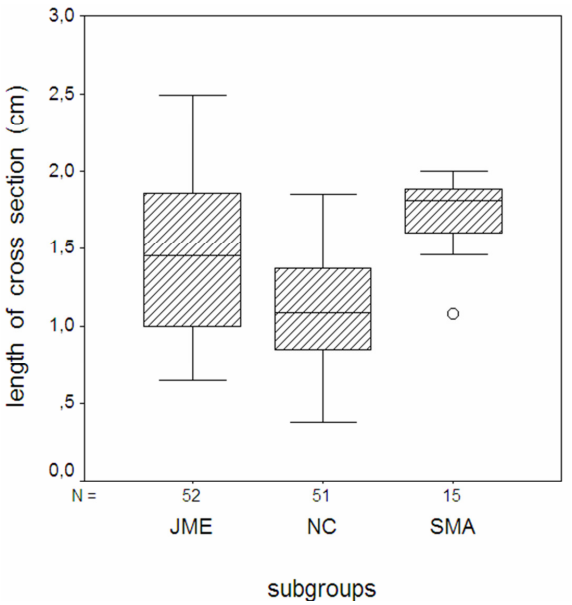


Figure 8.1 The box-plots of lengths of cross-sections for JME, NC and SMA groups (N represents number of sessions per group).

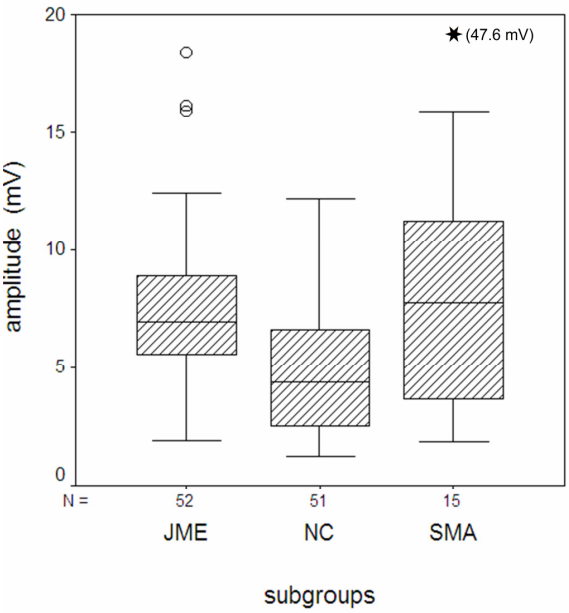


Figure 8.2 The box-plots of maximum amplitudes for JME, NC and SMA groups (N represents number of sessions per group).

When JME and NC groups were compared in terms of number of motor units being studied by using Student's t-test with 95% CI, an extremely significant difference was found between the lengths of cross-section and the maximum amplitudes of both groups.(p<0.01). The comparison between JME and SMA group exhibited a marginal significance for lengths of cross-sections (p<0.1), but the maximum amplitudes were not significantly different (p>0.1). This suggested that the JME and SMA groups are similar. When NC and SMA groups were compared, the difference between the lengths of cross-section of either group was found extremely significant (p<0.01) and that of the maximum amplitudes were found very significant (p>0.01). This is expected due to the reinnervation usually occurring in SMA.

One-way ANOVA indicated that the overall differences both in terms of length of cross-section and maximum amplitude between three subject groups was extremely significant (p<0.001). Hence, Tukey's HSD test can be performed all of these three groups at the same time in order to minimize Type I error that will be folded by multiple t-test application.

When JME group was compared with other groups in terms of length of cross-section, the difference between JME and NC groups was extremely significant (p<0.001). However, JME group was not significantly different from SMA group (p>0.1). When JME group was compared with other groups in terms of maximum amplitude, NC was significantly different from both of other groups (p<0.01). For SMA group, the difference in the length of cross-section with JME was not significant (p>0.1). However, there was an extremely significant difference between SMA and NC groups (p<0.001).

When NC group was compared with others, the difference with JME was marginally significant (p<0.1). However, it was extremely significant with SMA (p<0.001) which is expected due to the reinnervation process in the course of SMA. In terms of maximum

amplitude, there was a significant difference between JME and NC groups (p>0.01), however, the difference was not significant between JME and SMA groups (p>0.1). NC was significantly different from JME, but it had extremely significant difference with SMA group (p<0.01).

SMA group was not significantly different from JME group (p>0.1), however, it had had extremely significant difference with NC group (p<0.01). As a result, JME and SMA groups seem similar to each other in terms of both variables. On the other hand, NC group is different from other groups in terms of both variables.

These groups were compared also in terms of subjects. The median values for both parameters were considered beside the mean values for each subject. This was achieved first by using Student's t-test in 95% CI.

When the mean length of cross-section and that of the median lengths of cross-section were compared between JME and NC groups, the difference was significant for both variables (p>0.01). The mean of maximum amplitudes was marginally significant between JME and NC groups (p<0.01). But the mean of median maximum amplitudes was significantly different between these groups (p>0.01).

When the JME and SMA groups were compared in terms of mean lengths of cross-section, mean of median lengths of cross-section, mean of maximum amplitudes and mean of the median maximum amplitudes there were not significantly different (p>0.1).

The mean of length of cross-section and the mean of median length of cross-section were extremely significant between NC and SMA group (p<0.01). However, the mean of

maximum amplitudes were not significant between these groups (p> 0.1). The mean of median maximum amplitude was found significantly different between these groups (p>0.01).

Since the distributions are not normal in terms of number of subjects the JME, NC, and SMA groups were also compared in pairs with a non-parametric test such as Mann-Whitney Test. The mean of the mean length of cross-section was not found significantly different between JME and NC groups (p>0.1). The difference in the mean of median of length of cross-section was found marginally significant between these groups (p<0.1). The mean of the mean maximum amplitude and the mean of the median maximum amplitudes were found significantly different between these groups (p>0.01).

For JME and SMA groups, the mean length of cross-section, the mean of the median lengths of cross-section, the mean of the maximum amplitudes and the mean of the median maximum amplitudes, all, were not found significantly different (p>0.1). For SMA and NC groups, the mean of the mean length of cross-section, the mean of the median lengths of cross-section were significantly different (p>0.01). However, the mean of the mean maximum amplitudes and the mean of the median maximum amplitudes, were not found significantly different (p>0.1).

One-way ANOVA indicated that the overall differences both in terms of the mean lengths of cross-section and the mean of the median lengths of cross-section, the mean of the mean maximum amplitudes and the mean of the median maximum amplitudes between three subject groups was significant (p>0.01).

All groups were again compared with Tukey's HSD test in terms of each subject. For the mean of the means lengths of cross-sections, JME group was not found significantly

different from other groups ($p>0.1$). NC and SMA groups were not also found significantly different from JME group ($p>0.1$). However, there was a significant difference between SMA and NC groups ($p>0.01$). For the mean of the median length of cross section amplitudes, there was no significant difference between JME and NC groups and JME and SMA groups ($p>0.1$). However, there was a marginally significant difference between SMA and NC groups ($p<0.1$).

For the mean of mean maximum amplitudes, there was a marginally significant difference between JME and NC groups and NC and SMA groups ($p<0.1$). SMA and JME groups were not significantly different ($p>0.1$). There was a significant difference between NC and SMA groups and NC and JME groups ($p>0.01$). For the mean of the median maximum amplitudes, there was a marginally significant difference between JME and NC groups ($p<0.1$). NC and SMA groups and JME and SMA groups were not found significantly different ($p>0.1$).

Since the number of subjects is less than 30, Kruskall-Wallis Method being non-parametric one-way ANOVA was used to compare three groups all together. The mean lengths of cross-section and the mean of the median lengths of cross-section, the mean of the mean maximum amplitudes and the mean of the median maximum amplitudes between three subject groups were significantly different ($p>0.01$).

As a result, the JME group is again similar to SMA group. However it is significantly different from NC group. This suggests again the presence of large motor units in JME group.

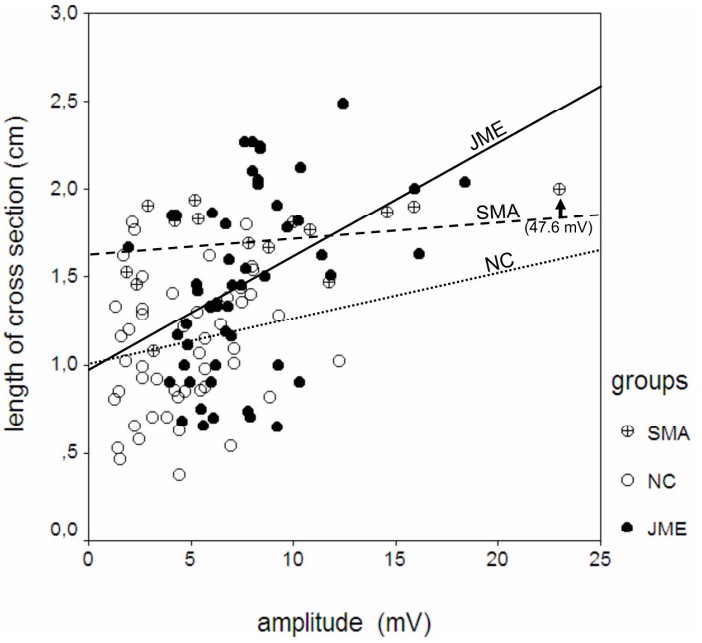


Figure 8.3 The correlation between the length of cross-section and the amplitude for JME, SMA and NC groups.

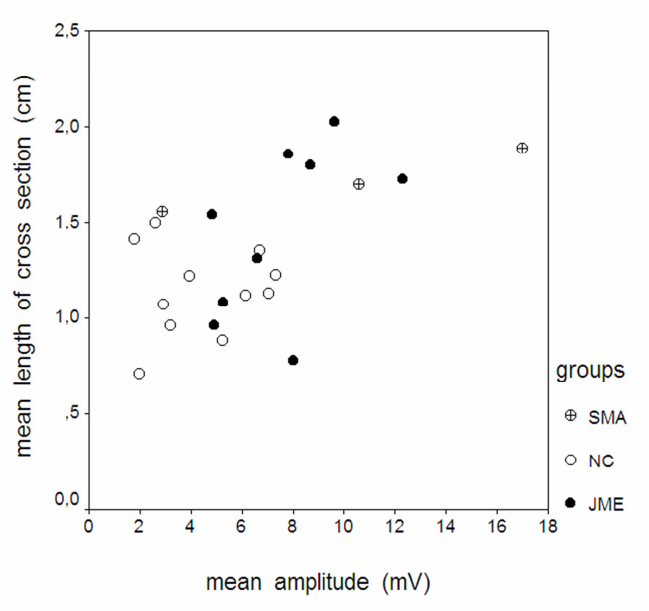


Figure 8.4 The scatter plot of the relationship between the mean lengths of cross-section and the mean maximum amplitudes.

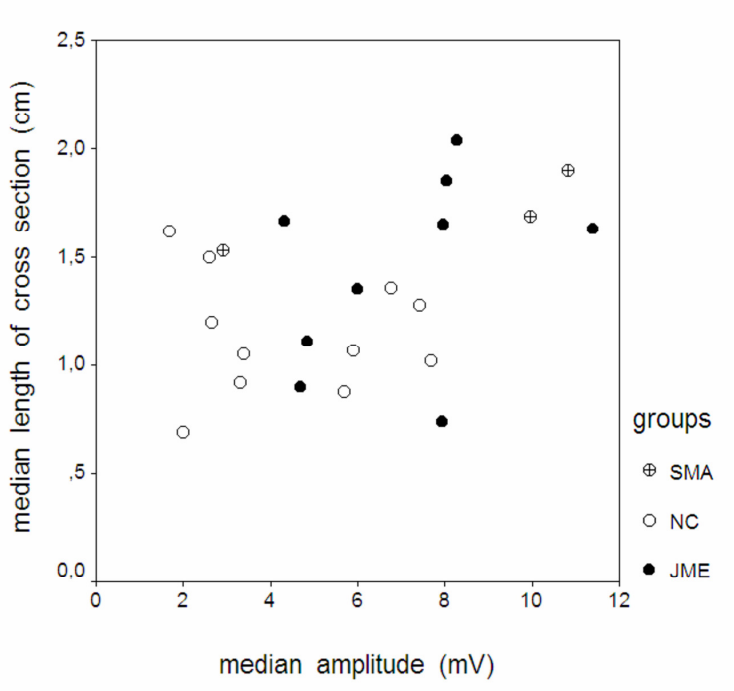


Figure 8.5 The scatter plot of the relationship between the mean lengths of cross-section and the mean maximum amplitudes.

The correlation between the maximum amplitude and the length of cross-section was also studied. This is shown in Figure 8.3 for all of the three groups when the number of motor units is considered. In JME group, a stronger correlation can be observed when compared to other groups. In case of individual subjects also, stronger correlation can be observed between mean lengths of cross-section (Figure 8.4) and mean maximum amplitude and median lengths of cross-section and median maximum amplitudes (Figure 8.5) in JME group compared to other groups. In correlation analysis, there was a very significant correlation between these variables in JME (r^2=0.142 and p>0.01). On the other hand, these parameters were not correlated in SMA (r^2=0.183 and p>0.1) and NC (r^2=0.032 and p>0.1) groups. In normal controls such a correlation is not expected. This is the same for SMA group also, because, the amplitude increases due to the reinnervation even the length of cross-section does not increase. In JME group, as the amplitude increases, the length of cross-section also increases. The increase in the

maximum amplitude with the increase in the length of cross-section was observed only in patients with JME. This may be due to the special configuration of MU territory in which the density of muscle fibers may be slightly increased than normal configuration.

In order to study the relationship of the age with the length of cross-section and the maximum amplitude, a correlation analysis was performed for JME, NC and SMA groups. For JME group, the relationships of the age with the mean length of cross-section (r^2=0.128, p>0.1), the median length of cross-section (r^2=0.143, p>0.1), the mean maximum amplitude (r^2=0.00, p>0.1) and the median maximum amplitude (r^2= 0.052, p>0.1) were not found as correlated. For SMA group also the relationships of the age with the mean length of cross-section (r^2=0.754, p>0.1), the median length of cross-section (r^2=0.753, p>0.1), the mean maximum amplitude (r^2=0.898, p>0.1) and the median maximum amplitude (r^2= 0.958, p>0.1) was not found as correlated. For NC group, the relationships of the age with the mean length of cross-section (r^2=0.084, p>0.1), the median length of cross-section (r^2=0.081, p>0.1) were not correlated, however, the mean maximum amplitude (r^2=0,476, p>0.01) and the median maximum amplitude (r^2= 0.498, p<0.01) were found as correlated.

9. CONCLUSION

The statistical analysis performed with the data acquired during this study demonstrates that the parameters such as length of cross-section of a motor unit territory and the maximum amplitude of the motor unit based on the temporal and spatial characteristics in JME patients are similar to those of SMA patients. The significant differences between the JME patients and the healthy volunteers have been also revealed. As a result, it can be stated that the preponderance of the large motor units in biceps brachialis muscles of the juvenile myoclonic epilepsy patients is apparent when compared to normal individuals.

The effects of age have not been shown on the amplitude and the length of cross-sections, except the amplitude for only normal subjects. Therefore, since this evidence may discard the role of progressive processes, it might be suggested that the presence of large motor units in JME is structural

Due to the nature of study, only subjectss with SMA as a neurogenic condition were included. Therefore, only the length of cross-section of MUs and the maximum amplitude of MUs were investigated among the parameters that can be monitored. Since another subject group with any myopathic condition was not required, other parameters were not taken into account in assessing the electrophysiologic cross-sections of MUs that were investigated in this study.

This study can be expanded by increasing the number of subjects in the future for further statistical analysis. The experimental setup can be improved by using more powerful computer system and by implementing several modifications into the interface software in order to use in other studies related to motor unit territory in patients with

other neuromuscular diseases. Besides, a subject group with a myopathic disorder can be included in order to have a complete data set for neurogenic, myopathic and normal conditions. Therefore, not only the length of cross-sections of MUs and the maximum amplitudes of MUs but also silent periods, fractions of MUs, polyphasic and complex portions of MUAPs can be also considered. As a result, the 3-D elctrophysiological of these conditions can be classified by means of pattern recognition algorithms and thus, by using artificial neural networks. This classification method may be an auxillary diagnostic tool in differentiating the neuromuscular disorders.

Current data can be used to study the role of hypertrophy in the preponderance of large MUs in either JME or SMA subjects. By performing Power Spectral Analysis (PSA), mean power frequency can be measured. Since the occurrence of the large MUs in JME is structural, it is expected that mean power frequency will lower in JME compared to SMA. Because the firing rate will be increased in SMA due to the reinnervation, the mean power frequency should be higher that that of signals acquired from JME subjects.

Inclusion body myositis (IBM) is an inflammatory myopathic condition that exhibits confusing electrophysiological findings. It can demonstrate evidence as a neurogenic condition in CNEMG where as, it can give findings as a myopathic condition with Macro-EMG. The absolute diagnosis can be made by biopsy. However, if the correct diagnosis cannot be established, the effective treatment for this disorder is not possible. Thus, the electrophysiological cross-section of the MUs of the subjects with IBM can be investigated by using the experimental setup of Scanning EMG built in this study. Due to the degeneration in muscle fibers in IBM, parameters that reflect the topographic alterations in MU such as silent periods, number of fractions of MUs can be investigated. Besides, maximum amplitude can also be measured for the comparison with other subject groups in order to clarify whether moypathy is more prominent or not. The subject may be grouped as acquired inflammatory disease including one group for IBM and one group for polymyositis (PM), one neurogenic disease including one SMA group, one normal

control group and one myopathic disorders including one muscular dystrophy group. The significance of differences between IBM group and other groups can be studied in terms of the parameters stated as above. Furthermore, it can be examined also whether Scanning EMG can be used as a diagnostic tool for IBM patients.

APPENDIX

Table A.1

Length of Cross-sections (in cm) of the Motor Units for JME group

(Si=Subjects; MUi=Motor Units)

	S1	S2	S3	S4	S5	S6	S7	S8	S9
MU1	2.23	2.24	1.82	0.90	1.78	0.90	1.86	1.19	1.00
MU2	2.12	2.27	2.00	1.00	1.35	1.23	1.91	1.16	0.90
MU3	2.49	1.45	1.63	0.68	0.66	1.17	2.27	1.85	0.71
MU4	1.45	2.10	1.62	1.33	1.42	1.11	2.05	1.67	0.75
MU5	1.33	1.60	1.55	0.90	1.33	1.00	2.03	1.85	0.74
MU6	1.46	1.50					2.04		0.70
MU7	1.80								0.65
MU8	1.51								

Table A.2

Length of Cross-sections (in cm) of the Motor Units for NC group

(Si=Subjects; MUi=Motor Units)

	S1	S2	S3	S4	S5	S6	S7	S8	S9	S10
MU1	0.71	1.30	1.50	1.02	1.81	0.85	0.54	1.41	0.99	0.86
MU2	1.15	1.40	1.02	1.80	1.77	1.44	1.07	0.63	0.58	1.22
MU3	1.23	1.36	1.09	0.82	1.33	1.01	0.82	1.20	0.47	1.16
MU4	0.86	1.38	0.93	0.71	1.62	1.54	1.56	1.29	0.81	0.92
MU5	0.88	1.33	1.85	1.32	0.53	1.28	1.62	0.85		0.66
MU6	0.38									
MU7	0.98									

Table A.3

Length of Cross-sections (in cm) of the Motor Units for SMA group

(Si=Subjects; MUi=Motor Units)

	S1	S2	S3
MU1	1.91	1.69	2.00
MU2	1.46	1.81	1.94
MU3	1.82	1.87	1.90
MU4	1.53	1.47	1.77
MU5	1.08	1.67	1.83

Table A.4

Maximum Amplitudes (in mV) of the Motor Units for JME group

(Si=Subjects; MUi=Motor Units)

	S1	**S2**	**S3**	**S4**	**S5**	**S6**	**S7**	**S8**	**S9**
MU1	8.43	8.39	10.27	3.97	9.71	6.00	6.05	6.71	9.25
MU2	10.37	8.03	15.96	4.68	6.25	4.83	9.20	6.97	10.32
MU3	12.45	7.02	16.17	4.58	5.64	4.37	7.63	4.32	7.93
MU4	7.50	8.03	11.39	6.25	5.34	4.84	8.29	1.94	5.49
MU5	6.81	6.86	7.72	4.98	6.00	6.20	8.29	4.17	7.78
MU6	5.28	8.64					18.40		6.10
MU7	6.71								9.20
MU8	11.84								

Table A.5

Maximum Amplitudes (in mV) of the Motor Units for NC group

(Si=Subjects; MUi=Motor Units)

	S1	**S2**	**S3**	**S4**	**S5**	**S6**	**S7**	**S8**	**S9**	**S10**
MU1	3.10	5.29	2.59	12.20	2.14	4.68	6.91	4.07	2.64	4.22
MU2	5.69	7.93	1.78	7.68	2.24	7.42	5.39	4.42	2.44	4.63
MU3	6.46	7.47	7.12	8.85	1.33	7.12	4.38	1.98	1.53	1.58
MU4	5.44	6.76	2.64	3.81	1.68	8.03	7.98	2.64	1.28	3.30
MU5	5.69	5.95	4.12	2.59	1.43	9.25	5.90	1.47		2.24
MU6	4.42									
MU7	5.69									

Table A.6

Maximum Amplitudes (in mV) of the Motor Units for SMA group

(Si=Subjects; MUi=Motor Units)

	S1	**S2**	**S3**
MU1	2.90	7.78	47.55
MU2	2.34	9.97	5.21
MU3	4.22	14.59	15.89
MU4	1.83	11.74	10.81
MU5	3.15	8.77	5.34

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