MUSCLE PHYSIOLOGY

(a) ***To describe the comparative physiology of skeletal, smooth, and cardiac muscle.***

(b) ***To describe the muscle spindle and Golgi organ and to explain their physiological roles.***

(c) ***To describe the physiology of the neuromuscular junction and its receptors.***

(d) ***To describe the mechanism of excitation-contraction coupling.***

(e) ***To describe the different types of skeletal muscle fibres (Ie. fast or slow).***

(f) ***To explain the concept of motor units.***

(g) ***To describe the monosynaptic stretch reflex.***

(h) ***To define single twitch, tetanus and Treppe effect, and explain their physiological basis.***

(i) ***To describe the relationship between muscle length and tension.***

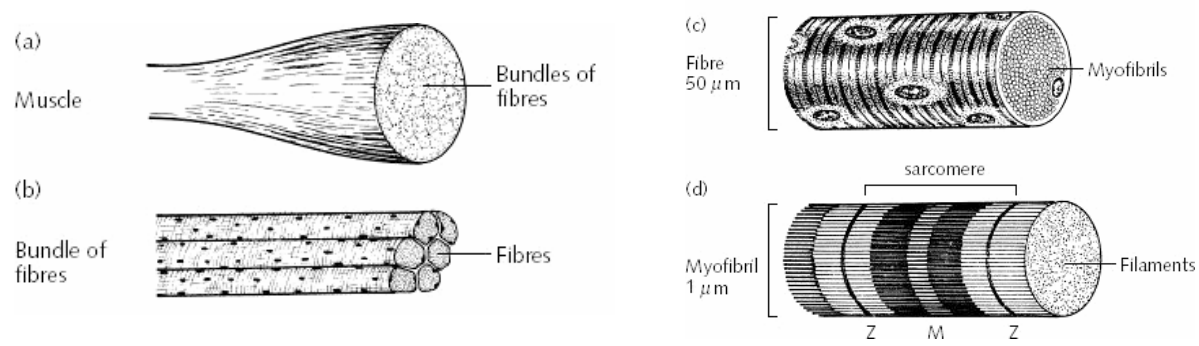
Skeletal Muscle:

(I) Anatomy of Skeletal Muscle:

"Muscle Fibre" → basic cell unit of skeletal muscle

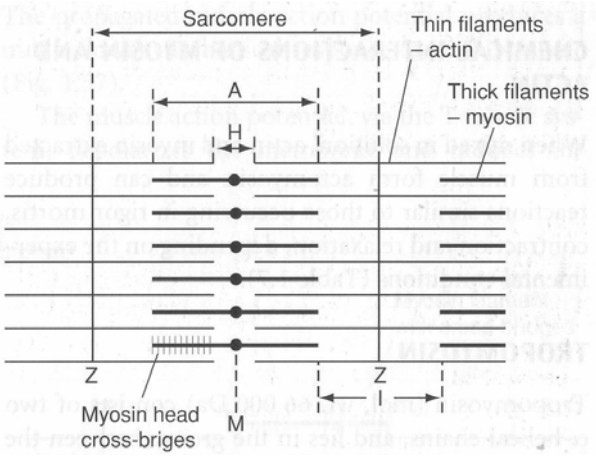
- ~10-100 um diameter and length
- Contains:
  - (i) Multiple nuclei (~100) on periphery of cell
  - (ii) Myofibril (consisting of several sarcomeres in series – See below)
  - (iii) Sarcoplasmic reticulum (SR) – Network of vesicular elements running longitudinally around myofibril → sequesters Ca^{2+} by Ca^{2+}/Mg^{2+}-dependent ATPase
  - (iv) T-tubules – Invaginates at regular places on myofibril (at A-I junction) and forms a triad with 2 lateral sacs of SR → permits muscle AP to be transmitted from the surface muscle membrane down to the SR, which leads to SR Ca^{2+} channel opening and muscle contraction
  - (v) Mitochondria
  - (vi) Cytoplasm
- "Striated" appearance → due to interdigitating actin (thin filament) and myosin (thick filament) within sarcomere

"Fascicle" → bundle of muscle fibres



"Sarcomere" → basic unit of muscle contraction within myofibril

- Consists of (i) contractile proteins (interdigitating actin (thin filament) and myosin (thick filament) → myosin heads form cross-bridges with actin filaments → gives muscle "striated" appearance) and (ii) elastic proteins
- Extends from Z to Z lines – contains 1x A-band and 2x half I-bands

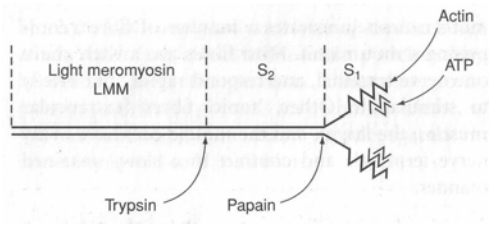


Z line – Actin anchors here
I band – Has actin filaments only
A band – Contains myosin and some actin
H band – Band of myosin only
M line – Joins myosin filaments together

Components of the sarcomere:

- (i) Myosin
  - Large protein that consists of:
    - (a) 1x long tail – 2 interwound α-helices with a flexible hinge (S2 segment)

- (b) 2x globular heads (S1 segment) – Connected to long tail via S2 segment. Each head has 1x heavy chain that binds 1x G-actin, and 2x short chains that bind ATP



Note – Myosin heads protrude out from thick filament radially, such that 6 actin filaments are arranged in a HEXAGONAL fashion around 1x myosin filament

  - 2x myosins fuse together at M-line via their long tails → forms "thick filament"
- (ii) Actin (thin filament)
  - Consists of 2x chains of F-actin (≈ double-strand cord) formed from the polymerisation of G-actin
- (iii) Tropomyosin
  - 2x α-helical chains that lie between 2x chains of actin polymers → under the influence of Troponin, it can acts to inhibit or permit myosin-actin interaction
- (iv) Troponin
  - Present with tropomyosin on thin filament at regular intervals (every 7 actin monomers)
  - Consists of 3 subunits – (a) T (binds tropomyosin), (b) I (inhibits myosin ATPase) and (c) C (binds Ca^{2+})
  - Binding to Ca^{2+} causes a conformation change in troponin-tropomyosin complex → allows actin to interact with myosin
- (v) Titin
  - Large elastic molecule that attaches myosin to Z-line → acts to return muscle to resting length

(II) <u>Types of Skeletal Muscle</u>:

(1) Extrafusal fibres
- Regular contractile fibres → supplied by α-motor neuron
- There are two types:
  - (a) Slow-type (tonic) muscle fibres:
    - Very uncommon in body (includes laryngeal and extra-ocular muscles)
    - Supplied by multiple "grape-like" nerve terminals that distribute over the whole muscle → repeated nerve stimulation produces a muscle AP throughout the muscle (Ie. muscle AP is not propagated from a single MEP) → generates a "tonic" contraction (slow and sustained)
  - (b) Fast-type (twitch) muscle fibres
    - Common in body → most muscles
    - Supplied by a single nerve terminal → single nerve stimulation produces a muscle AP that propagates from the MEP → generates a "twitch" contraction (rapid and brief)
- Of the fast-type (twitch) muscle fibres, there are three types:

	Fast fatigable fibres	Slow fatigue-resistant fibres	Fast fatigue-resistant fibres
Also known as	Type IIB, white muscle or fast glycolytic fibre	Type I, red muscle or slow oxidative fibre	Type IIA, red muscle or fast oxidative fibre
Main location	White muscle	Red muscle	Red muscle (but small component cf. type I)
Function	Fine/precise movements (Ie. hand	Strong, sustained contractions (Ie.	Fine/precise movements

	muscles)	postural muscles)	
Onset of twitch	Fast	Slow	Fast
Duration of twitch	Short (7.5-10 msec)	Sustained (100 msec)	Short (7.5-10 msec)
Fatigability	++++	+	++
Energy generation and consumption	- Anaerobic metabolism via glycolysis → ↑ glycolytic enzymes, ↓ mitochondria and myoglobin content - Fast myosin ATPase and high SR Ca^{2+} pumping capacity - Minimal ATP made but more ATP consumed rapidly → easily fatigues	- Aerobic metabolism via [O] phosphorylation → ↑ mitochondria and myoglobin content, and ↓ glycolytic enzymes - Slow myosin ATPase and low SR Ca^{2+} pumping capacity - More ATP made but less ATP consumed → fatigue-resistant	- Aerobic metabolism via [O] phosphorylation → ↑ mitochondria and myoglobin content, and ↓ glycolytic enzymes - Slow myosin ATPase and low SR Ca^{2+} pumping capacity - More ATP made but less ATP consumed → fatigue-resistant
Note – "Red" muscles contain ↑ [Mb] which gives them its red appearance (cf. "White" muscle which lacks Mb)			

(2) Intrafusal fibres – Muscle spindles that lie parallel to extrafusal fibres → supplied by γ-motor neuron

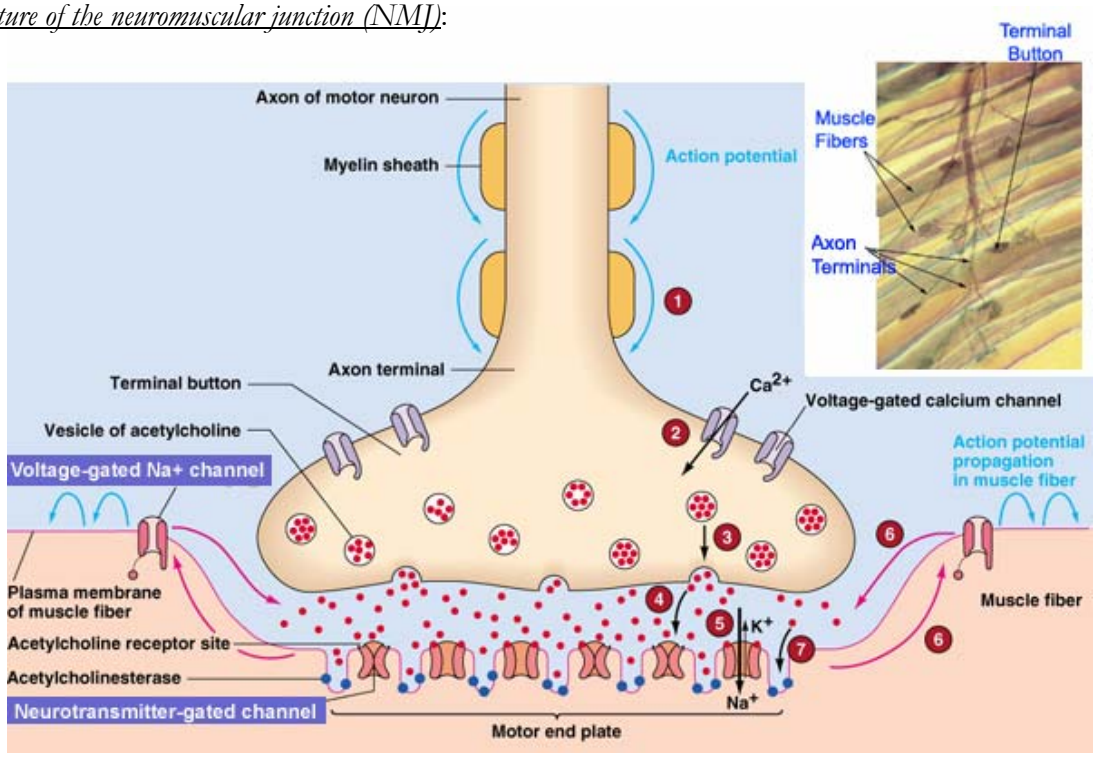
(III) Motor Unit:

- Consists of a single α-motor neuron and all the extrafusal skeletal muscle fibres it supplies
- Considered the "functional unit of contraction" → b/c it produces the smallest amount of muscle contraction in response to stimulation of the α-motor neuron

> Note – # of muscle fibres in a motor unit varies → muscles used for fine movement (Ie. hand muscles) have small motor units (few fibres per motor neuron), while muscles used for coarse movements (Ie. postural muscles) have larger motor units (several fibres per motor neuron)

(IV) Neuromuscular Transmission:

Structure of the neuromuscular junction (NMJ):



(1) "Nerve terminal" of the motor neuron → contains:
- (i) Synaptic vesicles – Found in cytoplasm and along the presynaptic membrane flanking a "dense bar" (forms an "Active zone" opposite to nAChR on MEP) → contain ACh stores that can be released into synaptic cleft upon depolarisation of the nerve terminal
- (ii) VG-Ca^{2+} channel – Permit Ca^{2+} influx in response to motor nerve AP → causes ↑ IC [Ca^{2+}] that mobilise synaptic vesicles for ACh release
- (iii) Prejunctional nAChR – Cause +ve feedback on ACh release when stimulated by ACh
- (iv) Mitochondria – Produces acetyl-CoA from pyruvate and PDH

(2) "Synaptic cleft":
- ~ 500 Angstrom wide
- Contains AChE (within the (i) junctional folds and (ii) extracellular material (basal lamina)) → rapidly hydrolyses ACh into choline and acetate

(3) "Motor end plate":
- Area of specialised muscle membrane that forms a series of folds
- Contains several ligand-gated nAChR (at the crests of these folds) → causes influx of cations (Na^+, K^+, Ca^{2+}, Mg^{2+}) when stimulated by ACh, leading to production of localised end-plate potentials

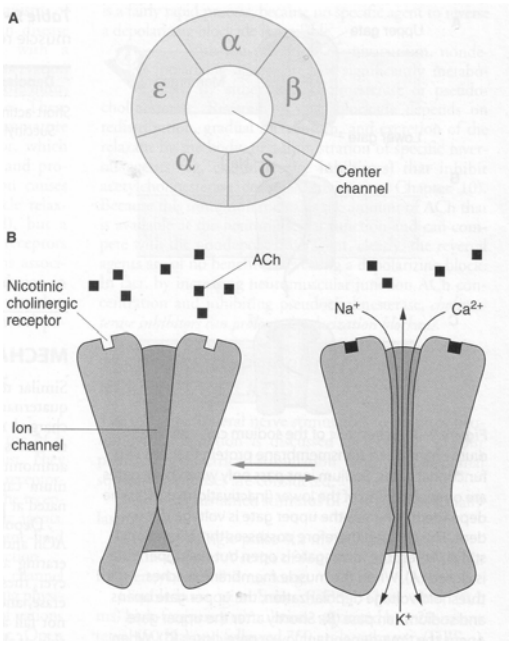
(4) Sarcolemma surrounding the MEP:
- Contains VG-Na^+ channels → Localised EPP at the MEP trigger opening of these channels, leading to production and propagation of a muscle AP → excitation-contraction coupling

Overview of the nicotinic receptor (nAChR):

The NMJ contains 3 types of nAChR:
- Two postsynaptic on skeletal muscle surface:
  - (i) Junctional nAChR
    - Located on the crest of the specialised post-junctional folds (directly opposite sites on nerve ending where ACh is released) → implicated in normal NMJ transmission
    - ~ 5 million receptors in the adult form (of which only 10% are needed to be stimulated to cause a muscle AP) – They can be upregulated (in response to SC injury, CVA, burns, prolonged immobility, MS, GBS, prolonged NMBD exposure) or downregulated (in response to myasthenia gravis, AChE overdose, OP poisoning)
  - (ii) Extrajunctional nAChR
    - Appear over entire post-junctional membrane (rather than confined to specialised motor end plate)
    - Present in small numbers in the foetal form as synthesis is suppressed by normal neural activity – They proliferate with loss of motor nerve activity due to a denervating event, but degenerate when neural activity returns
- Presynaptic on nerve ending:
  - Prejunctional nAChR
    - Specialised nAChR that differs structurally and functionally cf. post-junctional nAChR
    - Main role is to influence presynaptic vesicular release of ACh – Stimulation of nAChR helps mobilise further ACh vesicles for release as part of a +ve feedback loop

Structure and function of post-junctional nAChR:

- Structure – Transmembrane ligand-gated ion channel → Pentameric structure (2x α, β, δ, γ or ε subunits – depending on adult/foetal subtype) with a central ion channel pore (permits transmembrane flow of Na^+, Ca^{2+}, K^+) and an extracellular receptor (binds ACh)
- Function – Simultaneous binding of the two α subunits by an ACh each causes a brief conformational change in the receptor → opens the central ion channel and permits Na^+ and Ca^{2+} influx and K^+ efflux along their electrochemical gradients → produces a localised membrane depolarisation (or end-plate potential), which can summate and trigger a skeletal muscle AP (via activation of perijunctional VG Na^+ channels) if $V_{THRESHOLD}$ of -50 mV is reached



Note: There are 2 subtypes of nAChR – (i) Foetal nAChR and (ii) Adult nAChR

- Structurally – Both pentameric and very similar except for a difference in a single subunit (ε vs γ) → Foetal nAChR contains 2x α, β, δ and a **γ**; Adult nAChR contains 2x α, β, δ and a ε
- Functionally – Foetal nAChR differs from adult nAChR in that when activated by ACh, it is associated with a more prolonged opened ion channel state (allowing Na^+, Ca^{2+}, K^+ flow across their electrochemical gradients), which has two implications – (i) A single quanta of ACh can elicit a muscle AP, and (ii) Greater release of K^+ from muscle (contributes to hyperkalaemic response to denervation)
- Foetal nAChR usually disappears and adult nAChR is expressed during synaptic maturation, HOWEVER with denervating conditions, foetal nAChR can be upregulated in extrajunctional areas of the skeletal muscle membrane

Structure and function of pre-junctional nAChR:

- Structure – Has a different pentameric structure cf. post-junctional nAChR
- Function – Altered chemical binding characteristics, ion channel selectively (Ie. Na^+ but not Ca^{2+}), and preferential blockade during high-frequency stimulation cf. post-junctional nAChR

Process of NMJ transmission:

Important to note – NMJ transmission in an amplification process where the motor nerve AP produces a larger muscle AP

(1) Acetylcholine (ACh) synthesis → occurs in the nerve cytoplasm

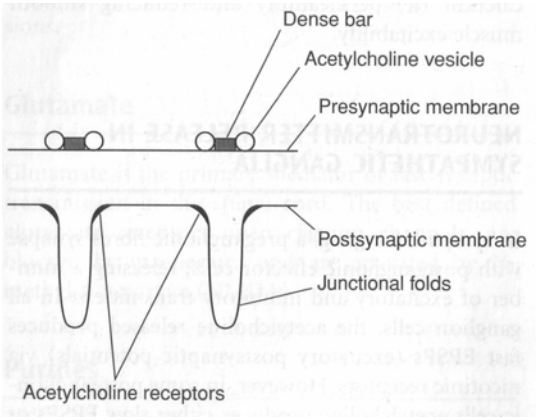
$$\text{Choline + Acetyl Coenzyme A} \xrightarrow{\text{Choline-O-Acetyl Transferase}} \text{Acetycholine}$$

Choline-O-Acetyl Transferase – Made in nerve cell body → transported to the axon
Acetyl Co-A – Made in mitochondria within nerve terminal from Pyruvate (using pyruvate DH)
Choline – Made by the liver or taken up from diet

(2) ACh storage:

- (i) Nerve terminal vesicles (80% – 1° store)

- Each vesicles contain – ACh (ATPase actively pumps ACh into the vesicle), ATP, Ca^{2+}, cholesterol, phospholipids, vesiculin
- Vesicles are found in (a) nerve terminal cytoplasm and (b) along the presynaptic membrane flanking a "Dense bar" (Nb. 2x vesicles line up on either side of the "dense bar" to form an "Active zone" opposite to nAChR on the MEP)
- Motor nerve AP causes IMMEDIATE release of this ACh store onto the MEP



- (ii) Stationary store (20%)
  - ACh is dissolved in the nerve terminal cytoplasm
  - Motor nerve AP causes SLOW and DELAYED release of this ACh store onto the MEP
- (iii) Surplus stores – Detected ONLY when IC AChEi is applied

(3) ACh release:
- (a) Spontaneous release:
  - Vesicles spontaneously and randomly fuse with the presynaptic membrane and empties ACh into the synaptic cleft via vesicular exocytosis INDEPENDENT of any motor nerve activity
  - Spontaneous and random quantal release of a single vesicle's content of ACh (fixed at 1500 molecules) produces a "Miniature end-plate potential" (MEPP) of constant amplitude (0.5-1 mV) → it is insufficient to depolarise the MEP to threshold, but has a role in maintaining an end-plate distribution of nAChR (Nb. denervation abolishes MEPPs, leading to spread of nAChR extrajunctionally!)

> 1 quantum = 1500 ACh molecules = 1x vesicle content = 0.5-1 1 mV = 1 MEPP

- (b) Motor nerve AP-evoked release:
  - Motor nerve AP stops at the last Node of Ranvier → produces local/electrotonic currents in the nerve terminal → opens VG-Ca^{2+} channels in nerve terminal membrane leading to Ca^{2+} influx
  - Ca^{2+} in the nerve terminal binds calmodulin → form Calci-Calmodulin complex → activates enzymes that change structural proteins on the vesicle and prejunctional membrane (Eg. synapsin)
  - This leads to exocytosis of 200 vesicles (~ 300k ACh) into the synaptic cleft and produces an end-plate potential at the MEP → the amount of ACh released with each motor nerve AP is 5-10x the minimal amount needed to depolarise the MEP to threshold potential and generate a muscle AP → this provides a wide safety margin to ensure effective NMJ transmission

> Note – Not every motor nerve AP causes vesicular exocytosis → in fact, repeated motor nerve AP causes fewer vesicles to be released per impulse

  - Prejunctional MP is restored to resting state by delayed opening of K^+ channels

(4) ACh function:
- (i) 1x ACh molecule binds to each of the α subunits of the post-synaptic nAChR (located mainly at the MEP crest of the junctional fold; but also at extrajunctional sites, esp with denervated muscle fibres) → this causes:
    - (i) Brief opening (~ 1 msec) of the channel associated with the receptor → leads to rapid flux of various cations across it (esp Na^+, K^+, Mg^{2+}, Ca^{2+})
    - (ii) Entry of these cations causes the MEP to depolarise from a RMP of -90 mV to "threshold potential" (-50 mV) → this generates a localised EPP that depolarises the surrounding muscle membrane (sarcolemma) → results in a propagated muscle AP that leads to muscle contraction via "excitation-contraction coupling"

	MEP potential	Muscle AP
Change in membrane ion permeability	↑ to Na^+, K^+, Mg^{2+}, Ca^{2+} simultaneously	↑ to Na^+, then K^+ later
Change in potential	Graded up to "threshold potential"	"All or none"
Propagated?	No	Yes
Refractory period?	No	Yes

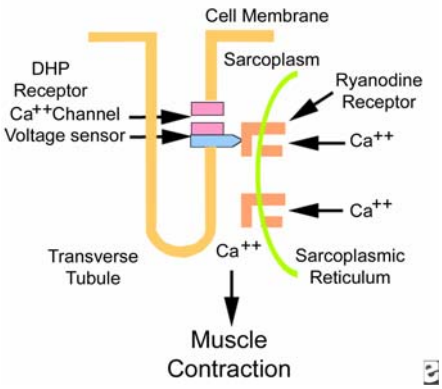
- (ii) Some ACh binds to prejunctional nAChR → this causes a +ve feedback on further presynaptic vesicular ACh release

(5) Inactivation of ACh:
- After binding to nAChR, ACh is then rapidly hydrolysed by AChE within the synaptic cleft to (i) choline and (ii) acetate → this allows terminates MEP stimulation and allows the MEP to return to its normal resting state
- Choline is recycled via reuptake into the nerve terminal where it is reused for synthesis of new ACh

> Aside – AChE is found in the synaptic cleft (within the (i) junctional folds and (ii) extracellular material (basal lamina)) → possesses 6 sites, each with anionic and esteratic components to attract and hydrolyse ACh into choline and acetate

(V) <u>Process of Muscle Contraction and Relaxation</u>:

(1) Excitation-contraction coupling:
- Describes the electro-mechanical process implicated in skeletal muscle contraction – involves motor nerve release of ACh at the NMJ → produces a muscle AP → leads to muscle contraction
- Process:
    - (i) Motor neuron terminal releases ACh at the NMJ → ACh binds nAChR at the MEP to produce an "End-plate potential" (EPP)
    - (ii) When EPP is at threshold (> -50 mV) → muscle AP is produced in the muscle membrane adjacent to the MEP
    - (iii) Muscle AP spreads across the muscle membrane, especially via the "Sarcotubular system":
        - AP travels via the T-tubules → triggers voltage-sensitive L-type Ca^{2+} channels (Dihydropyridine receptors) → causes – (a) Ca^{2+} influx into the cytoplasm, and (b) A "charge movement" across into the SR membrane
        - "Charge movement" opens SR Ca^{2+}-release channels (Ryanodine receptor) → results in Ca^{2+} release from the SR → ↑ $[Ca^{2+}]_{IC}$ from 0.1 uM at rest to 10 uM



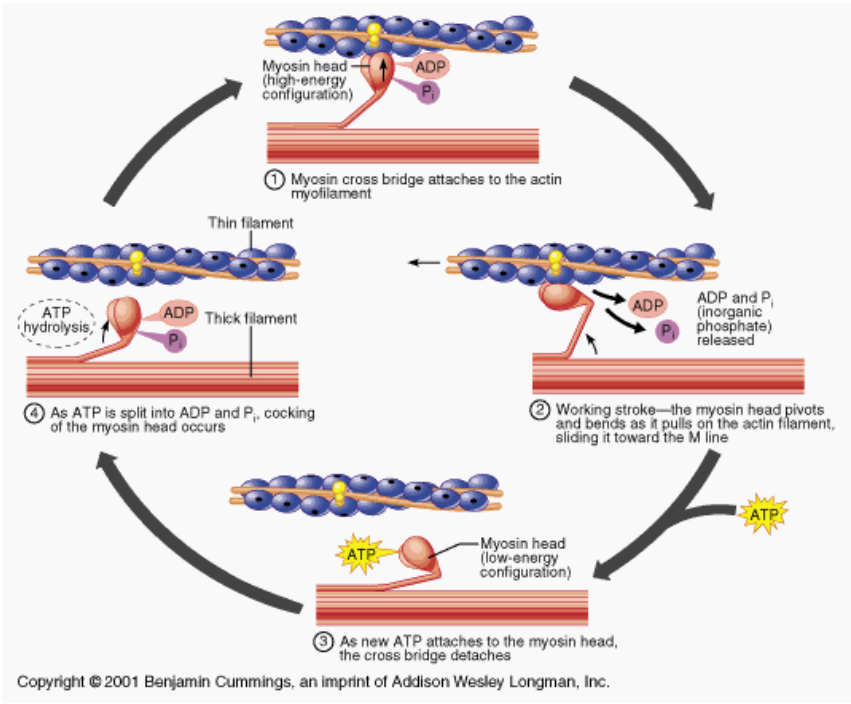
- (iv) Ca^{2+} interacts with Troponin-C → produces a conformational change that results in a shift in Tropomyosin → exposes actin to cross-bridge with myosin
- (v) Actin exposure removes Troponin I inhibition of Myosin ATPase → allows hydrolysis of myosin-bound ATP, which produces potential energy needed for the free myosin head to swing up and cross-bridge with a new actin molecule
- (vi) Muscle contraction then proceeds as per the "Sliding-Filament Theory"

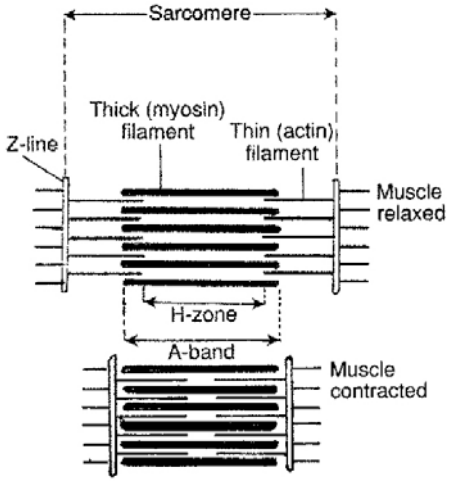
(2) Sliding filament theory:

- Describes the mechanical process of skeletal muscle contraction – involves repeated cycles of (a) actin-myosin head cross-bridge attachment, (b) pulling of actin fibres by the myosin head towards the centre of the sarcomere (causing thick and thin filaments to slide along each other), and then (c) detachment of actin-myosin head cross bridging
- Process:
  - (i) Actin+ myosin → produces rigor (stiffness)
  - (ii) Binding of ATP to myosin head of the actin-myosin complex releases actin from myosin → relaxation
  - (iii) ATP hydrolysis (to ADP + P_i) by myosin's ATPase → generates potential energy that causes the free myosin head to swing up and attach to a new actin molecule

> Note – Myosin ATPase is active ONLY when Troponin I-mediated inhibition of the ATPase is removed when actin is exposed → see "Excitation-contraction coupling" above

  - (iv) P_i is released causing a large burst of energy that results in the pushing of actin towards the centre of the sarcomere by the myosin head → "Power Stroke"





Important to note – Thick and thin fibres slide along each other, with myosin head cross-bridges pulling actin fibres towards the M-line → this causes:
- (a) Z-line to move together
- (b) I-bands to become smaller
- (c) A-bands to remain the same size

(3) Muscle relaxation → occurs as:
- (i) AChE metabolises ACh in the NMJ → ↓ nAChR activation at MEP → ↓ EPP and muscle AP generation
- (ii) SR actively sequesters Ca^{2+} (via Ca^{2+}-Mg^{2+} ATPase) → ↓ IC [Ca^{2+}] → Ca^{2+} removed from troponin C → tropomyosin mediated inhibition of actin-myosin interaction returns
- (iii) Elastic components of the sarcomere causing the muscle to return to its normal length

(VI) <u>Energy Source for Muscle Contraction</u>:
- Energy is needed for muscle contraction AND relaxation as it is required for:
  - (1) Formation of actin-myosin crossbridges and pulling to shorten muscle length in muscle contraction (via Myosin ATPase as per the "Sliding-filament theory")
  - (2) Muscle relaxation by – (i) Sequestration of Ca^{2+} in the SR (via Ca^{2+}-Mg^{2+} ATPase), and (ii) Restoration of Na^+/K^+ balance after muscle AP (via Na^+/K^+ ATPase)
- ATP is the ONLY energy source that contractile proteins in muscle can use, and ATP store in muscle is VERY limited (Ie. ATP in the muscle lasts for 8 twitches ONLY!) → thus, ATP is needed to be constantly generated
- The method of ATP generation depends on the level of activity present:
  - (1) Creatinine phosphate store
    - With intense exercise, ATP levels deplete quickly and is rapidly regenerated using creatinine phosphate stores (using Creatinine phosphokinase):

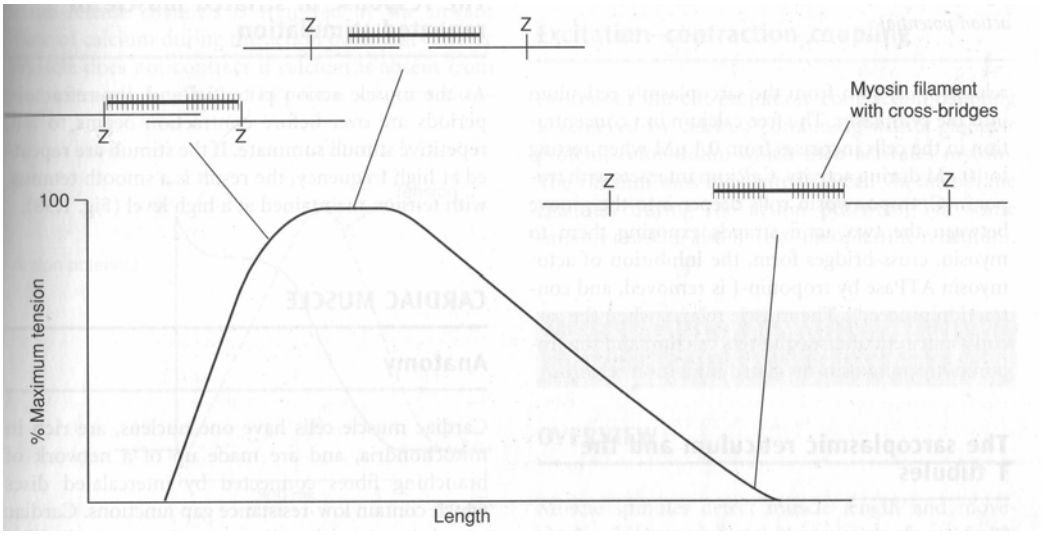
      $$\text{ADP + Creatine phosphate} \xrightarrow{\text{Creatine phosphokinase}} \text{ATP + creatinine}$$

    - Creatinine phosphate stores are replenished at rest by reversing the above reaction by aerobic/anaerobic generation of ATP
  - (2) Aerobic generation of ATP:
    - (a) Glucose (and Glycogen stores) – ATP is produced during exercise/activity by substrate phosphorylation (via glycolysis) and oxidative phosphorylation (via Kreb's cycle and the ETC)
    - (b) Fat – β-oxidation of fat produces ATP during light exercise and at rest
  - (3) Anaerobic generation of ATP:
    - When O_2 supplies are insufficient, ATP is produced in smaller amounts from the anaerobic glycolysis of glucose → produces lactic acid

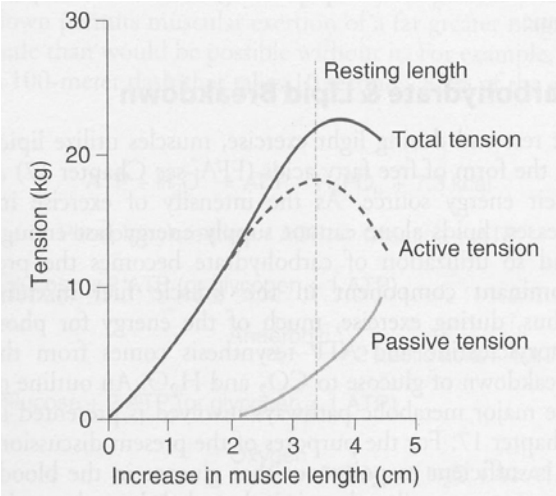
(VII) <u>Overview of Muscle Mechanics</u>:

(1) *"Length-tension relationship" of muscle*:

- The "active tension" developed during isometric muscle contraction depends on the initial muscle length → this is b/c the degree of overlapping actin-myosin filaments in the sarcomere determines the # of actin-myosin head cross bridges formed that can be used to develop muscle tension



- This relationship demonstrates that:
  - (i) Maximal "active tension" occurs at the muscle's "resting length" (which is where most skeletal muscle is arranged) → provides optimal degree of actin-myosin filament cross-bridging to provide maximal active tension
  - (ii) "Active tension" decreases with:
    - (a) Excess shortening of muscle fibre from its "resting length" → produces excess actin-myosin filament cross-bridging → ↓↓↓ distance the thin filament can move
    - (b) Excess stretching of muscle fibre from its "resting length" → produces minimal actin-myosin filament cross-bridging → ↓↓↓ distance the thin filament can move



This relationship is demonstrated experimentally by measuring the "passive" and "total" tension in skeletal muscle at various lengths:

- "Total tension" – tension developed when muscle contracts isometrically over a range of muscle lengths
- "Passive tension" – tension developed over a range of muscle lengths when it is not stimulated
- "Active tension" (or tension generated by "Muscle contractile elements") – difference between these two curves → muscle length at which active tension is maximal = "Resting length"

Important to note – Muscle tension can also be ↑↑↑ by:

- (a) Recruitment of multiple motor units
- (b) ↑ AP frequency to the motor units → causes "summation" of "muscle twitches", such that at a high frequency of motor nerve AP, a high level of tension ("Tetanus") is achieved (See below)

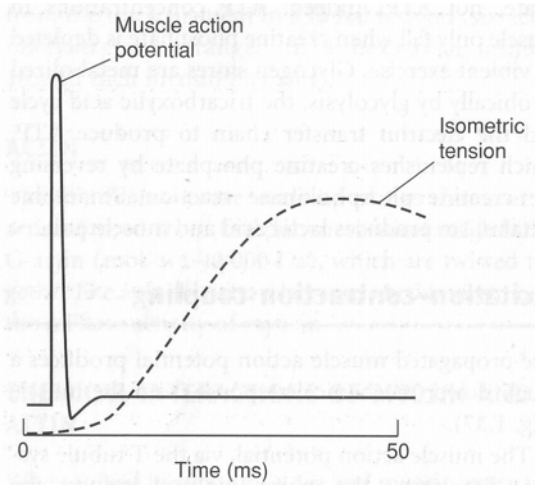
(2) *Velocity of muscle contraction*:

- (i) Velocity of muscle contraction varies inversely with the load on the muscle
- (ii) At a given load, velocity of muscle contraction is maximal at its "Resting length" (it declines at any other length)
- (iii) Velocity of muscle contraction depends on fibre type (fast vs slow)

(3) *Single twitch, Tetanus, and Treppe effect*:

Single muscle twitch:

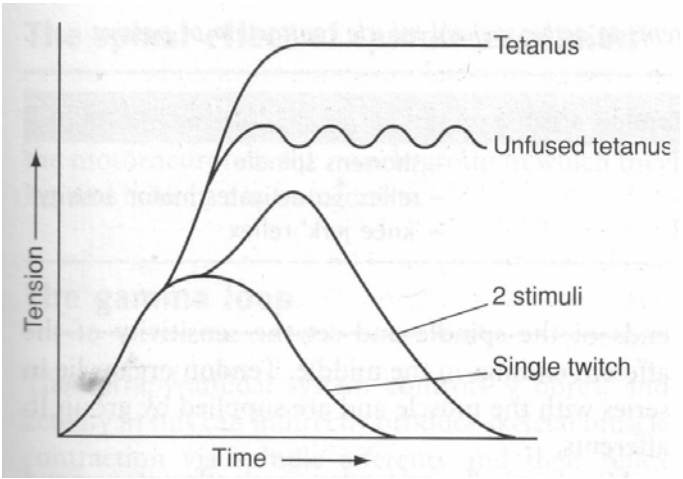
- Defined as one contraction-relaxation response to a single AP in a muscle fibre
- Physiological mechanism:
  - Single motor nerve AP stimulus produces a brief muscle AP → lasts 1-3 msec
  - This muscle AP then produces a slower and longer mechanical response in muscle (excitation-contraction coupling and sliding filament theory) → muscle contraction-relaxation cycle lasts b/t 7.5 to 100 msec (depending on type of fibre)



Important to note – Duration of muscle AP (and its refractory period) is LESS than the process of muscle contraction and relaxation

Tetanus:

- Defined as a state where high frequency motor nerve stimuli cause summation of individual muscle twitches into a continuous contraction that constantly maintains a high level of muscular tension → tension developed is much 3-4X than that of a single twitch
- There are two types:
  - "Complete tetany" – Absence of relaxation between summated stimuli → produces a sustained level of contraction with high level of muscular tension
  - "Incomplete tetany" – Periods of incomplete relaxation between summated stimuli → produces fluctuating level of contraction and muscular tension



- Physiological mechanism:
  - Motor nerve stimulation of muscle tissue is rapidly repeated above a "critical frequency" → causes repeated depolarisation of muscle tissue and generation of rapidly repeated muscle APs

> Note – "Critical Frequency" depends on the muscle fibre's single twitch duration → this is b/c twitch summation (which is required for tetany) only occurs IF the proceeding motor nerve and muscle AP occur PRIOR to the end of the muscle fibre's contraction-relaxation cycle (Ie. twitch duration = 10 msec → $f_{CRITICAL}$ = 100/s for twitch summation to occur)

  - Muscle contraction-relaxation cycle is a delayed and prolonged (7.5-100 msec) event cf. a muscle AP (1-3 msec) and muscle contractile elements do NOT have a refractory period → thus, rapidly repeated muscle APs result in activation of the muscle contractile mechanism prior to it completing muscle relaxation (Ie. repeated muscle AP → repeated Ca^{2+} release from SR before it can resequester it → leads to ↑ myoplasmic [Ca^{2+}] and ↑ actin-myosin cross-bridge formation → further muscle contraction)
  - This leads to "summation" of the contractile response (or single twitch) to the pre-existing level of muscle contraction → results in ↑ muscle tension
  - Repeated summation of these contractile responses (or twitches) forms a continuous contraction with a high level of muscular tension → produces "tetany"
- Important features of tetany to note:
  - It is a high-energy process (uses lots of ATP) due to (i) ongoing muscle depolarisation, and (ii) opposing action of Ca^{2+} ATPase in the SR
  - It ceases when (i) Muscle fatigue occurs (Ie. ATP depletion, local lactic acidosis), or (ii) Repetitive muscle depolarisation ceases

> Important to note – Tetany (and twitch summation) does NOT occur in cardiac muscle → this b/c the duration of cardiac muscle AP (and its refractory period) is nearly as long as the process of muscle contraction-relaxation

Treppe Effect (aka. "Bowditch Effect"):
- Observed in:
  - (1) Skeletal muscle – A series of motor nerve stimuli delivered to the muscle at a frequency just below "Critical frequency" required for tetany → produces a stepwise ↑ in tension developed during each twitch, such that after several contractions, a uniform tension per contraction is reached
  - (2) Cardiac muscle – Acts as an autoregulatory mechanism whereby a ↑ in heart rate leads to a stepwise ↑ in myocardial contractile force
- Physiological mechanism:
  - ↑ in muscle contractile tension is due to ↑ myoplasmic [Ca^{2+}], which binds troponin C and induces ↑ actin-myosin cross-bridging
  - This ↑ in myoplasmic [Ca^{2+}] is result of:
    - (i) Insufficient rate of Ca^{2+} sequestration in the SR
    - (ii) Impaired ability of membrane-bound Na^+/Ca^{2+} exchanger to extrude Ca^{2+} extracellularly (specific to cardiac muscle only) – ↑ HR causes (a) ↓ diastolic time → compromises time for exchanger to function, and (b) Na^+/K^+ ATPase to be unable to maintain a low IC [Na^+] for exchanger to function

(4) *Types of muscle contraction*:
- (i) Isotonic contraction ("same tension") – Creates force AND moves a load across a distance, thus work is done → there are two types:
  - (a) Concentric – Muscle shortens as constant tension applied → +ve work done
  - (b) Eccentric – Muscle lengthens despite a constant tension applied → –ve work done (Nb. damaging to muscle)
- (ii) Isometric contraction ("same length") – Creates force WITHOUT moving a load, thus no work is done

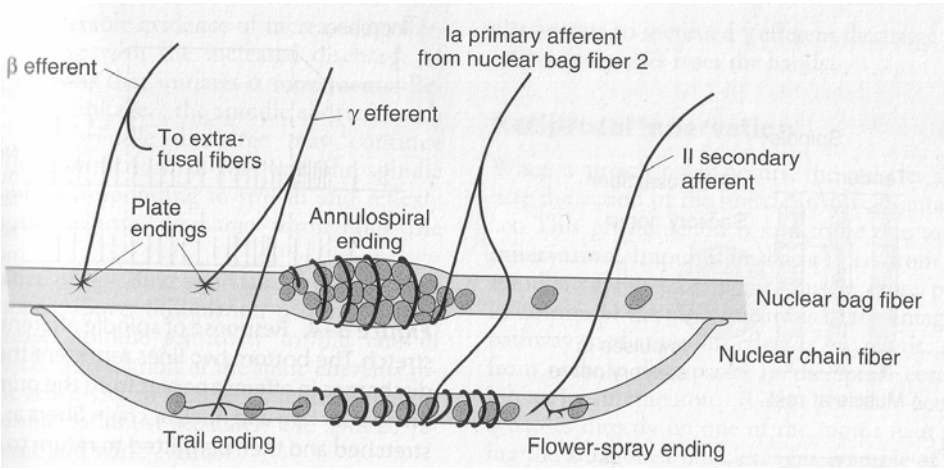
- Sarcomeres contract BUT elastic elements in series to the sarcomeres stretch (Eg. tendons, contractile filaments, cytoskeletal proteins), thus muscle length remains the SAME

(VIII) Muscle Spindle and Golgi Tendon Organ:

(1) Muscle spindle:

Structure of muscle spindle:

- Each muscle spindle consists of up to 10 "intrafusal" muscle fibres enclosed in a connective tissue capsule, and is arranged in PARALLEL to "extrafusal" fibres
- There are two types of intrafusal fibres within a spindle:
  - (i) Nuclear bag fibre (contains many nuclei in a dilated central area)
  - (ii) Nuclear chain fibre (thinner, shorter and more cylindrical)
- These intrafusal fibres have nerve connections:
  - (i) Afferent connections
    - Group Ia primary afferents – Annulospiral ending of nerves wrap that around the middle parts of the nuclear bag and nuclear chain fibres
    - Group II secondary afferents – Flower-spray endings of nerves that terminate towards the end of nuclear chain fibres only
  - (ii) Efferent connections
    - γ motor afferent nerve – Terminate on ends of nuclear bag and chain fibres
    - β motor afferent nerve - Terminate on end of nuclear bag fibre, and also on extrafusal fibres



Function of muscle spindle:

- Involved in the reflex control of – (i) Muscle length (static control), and (ii) Rate of change in muscle length (dynamic control)

> Important to note:
> - Output from nuclear chain fibres via Group II secondary afferents detect muscle length only → "static response"
> - Output from nuclear bag fibres via Group Ia primary afferents detect both muscle length AND the rate of change in muscle length → "dynamic response"

- Plays an role in maintaining muscle length via the "stretch reflex" in response to passive muscle stretching (See "monosynaptic spinal cord reflex" below)

Changes in muscle spindle during muscle contraction: a-γ linkage

- Muscle contraction causes ↓ tension of the associated muscle spindles and ↓ rate of impulses in their afferent nerves → this ↓ the sensitivity of the muscle spindle in the contracted muscle to detect muscle length and the rate of change in muscle length
- "α-γ linkage" prevents this and maintains muscle spindle function as follows:
    - α motor nerve activity causing muscle contraction causes concurrent stimulation of the γ motor neuron supplying the muscle spindles associated with the contracting muscle → this causes the muscle spindle ends to contract in unison with the contracting muscle
    - This shortens the length of the muscle spindle and ↑↑↑ it tension in association with the contracted muscle → thus maintains the sensitivity of the muscle spindle sensory receptors to muscle length and the rate of change in muscle length

Extrapyramidal control of muscle spindle: Gamma loop

- Muscle spindle activity can be influenced by the extrapyramidal system to indirectly produce skeletal muscle contraction via the "Gamma Loop" as follows:
    - (a) Extrapyramidal system controls γ fibres that cause intrafusal fibres to contract
    - (b) This in effect stimulates muscle spindle afferents that causes a spinal-mediated reflex activation of α fibres leading to extrafusal fibre contraction

(2) Golgi tendon organ (GTB):

- Structure – GTBs are a netlike collection of sensory receptors (supplied by group Ib afferent nerves) found in tendons at the end of muscle that lie in SERIES with the extrafusal muscle fibres
- Function – Detect the (i) rate and (ii) force of muscle contraction → plays a role in preventing muscle damage caused by excess active muscle contraction by retarding the force of muscle contraction via an inhibitory spinal reflex loop (See "monosynaptic spinal cord reflex" below) when the extrafusal fibres contract too strongly

(IX) Monosynaptic Stretch Reflex:

Important to note:

"Reflex" → automatic or involuntary stereotype response to a stimulus mediated by a "reflex arc"

"Reflex arc" consists of:

- (1) Receptor – A stimulus generates a "Receptor potential" whose magnitude is proportional to the stimulus strength
- (2) Afferent neuron – It enters into the central integrating station (Ie. via the dorsal roots, cranial nerves or autonomic nerves to cell bodies in the DRG, CN ganglia, or ANS ganglia, respectively) → "receptor potential" generates an AP in the afferent nerve, with the frequency of AP being proportional to the size of the "receptor potential"
- (3) Central integrating station – Found in the CNS (brain or spinal cord) or in the ANS ganglia → responses are graded by EPSPs and IPSPs at synaptic junctions connecting the afferent neuron with an interneurons or motor neuron
- (4) Efferent neuron – Often a motorneuron to the effector organ that leaves the central integrating station (Ie. via ventral roots, motor CN or autonomic nerves)
- (5) Effector – Efferent nerve AP produces a graded response in the effector

Reflexes can be classified by the number of neurons in the reflex pathway:

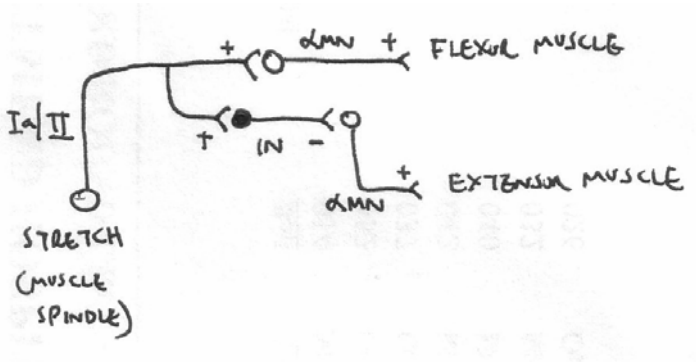
- (1) Monosynaptic – Single synapse between afferent and efferent neurons
- (2) Polysynaptic – One or more interneurons interposed between the afferent and efferent neurons

Muscle spindle and GTB play a vital role in monitoring and maintaining muscle movement via mono- and polysynaptic spinal cord stretch reflexes:

- (i) Muscle spindle → maintain muscle length in response to passive stretching of the muscle by reflexly contracting the muscle being stretched
- (ii) GTB → prevent muscle damage in response to excess active muscle contraction by reflexly minimising the force of muscle contraction

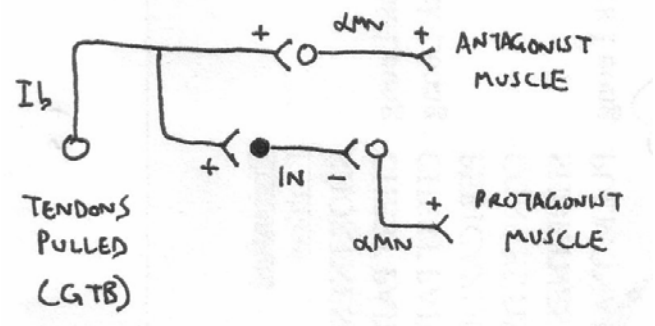
Muscle spindle (stretch) reflex (Eg. Patellar tendon (knee jerk) reflex):

- (i) Passive stretching of muscle → stimulates associated muscle spindles → send sensory output via Group Ia and II afferents → induces two spinal reflex arcs:
    - (a) Monosynaptic reflex arc – The afferent nerves synapse onto α motorneurons in the anterior horn of the SC that supply the same muscle group in which the muscle spindles lie → stimulate them to contract via release of excitatory NTs (Ie. glutamate) → maintains muscle length of the stretched muscle
    - (b) Reciprocal innervation – The afferent nerves also synapse onto inhibitory spinal interneurons → stimulate them via release of excitatory NTs (Ie. glutamate) → causes interneurons to inhibit α motor neurons supplying the antagonist muscle groups via release of inhibitory NTs (Ie. glycine) → inhibits their contraction, thus facilitating muscle length maintenance of the stretched muscle
- (ii) Once muscle length of the stretched muscle is restored, the muscle spindle is no longer stretched → thus the activity of the afferent nerve output and its triggered reflex arcs decrease back to its resting state



GTB (inverse stretch) reflex:

- (i) Excess active contraction of extrafusal fibres → pulls tendons → stimulates GTBs
- (ii) This induces an "Inverse Stretch Reflex" via Group Ib afferent nerves from the GTB → lead to concurrent activation of:
    - (a) Inhibitory spinal interneurons that inhibit α motor neurons supplying the same muscle group
    - (b) α motor neurons supplying the antagonist muscle group
- (iii) This weakens muscle contraction and removes the stimulation of the GTBs



Smooth Muscle:

(I) Anatomy of smooth muscle:

- Single nucleus
- Smaller than skeletal muscle (4 um diameter and ~ 400 um long)
- Contractile elements:
  - Actin/myosin extend diagnonally around cell → LACKS striated appearance
  - Have fewer myosin filaments → but they are longer and covered with more heads
  - Contain "Dense bodies" (rather than Z-lines) → attach actin to SM membrane
  - Lack troponin (but have tropomyosin)
- Have fewer sarcoplasmic reticula (poorly developed) and mitochondria (more reliant on glycolysis) than skeletal muscle
- Interconnected by "Gap junctions"

(II) Types of smooth muscle:

- (1) Visceral (single unit) smooth muscle
  - Found mainly in hollow viscera (Eg. vessels, gut, uterus, ureters)
  - Consists of large sheets of SM connected by Gap junctions (allows electrical contractile signals to pass through them)
  - Spontaneously depolarise ("Autorhythmic") due to an inward current via Ca^{2+} channels:
    - (a) Slow wave potentials – Spontaneous, alternating hyperpolarising and depolarising swings in membrane potential that produce an AP only if threshold is reached
    - (b) Pacemaker potentials – Membrane potential gradually depolarises to threshold on its own due to passive ionic fluxes across the membrane. This leads to a cyclical pattern in AP generation
  - Activity of smooth muscle can be varied by:
    - (a) Stretching of SM – Opens up Ca^{2+} channels
    - (b) ANS innervation – Modifies the degree of autorhythmicity
    - (c) Hormonal/paracrine agents – Modifies degree of autorhythmicity also

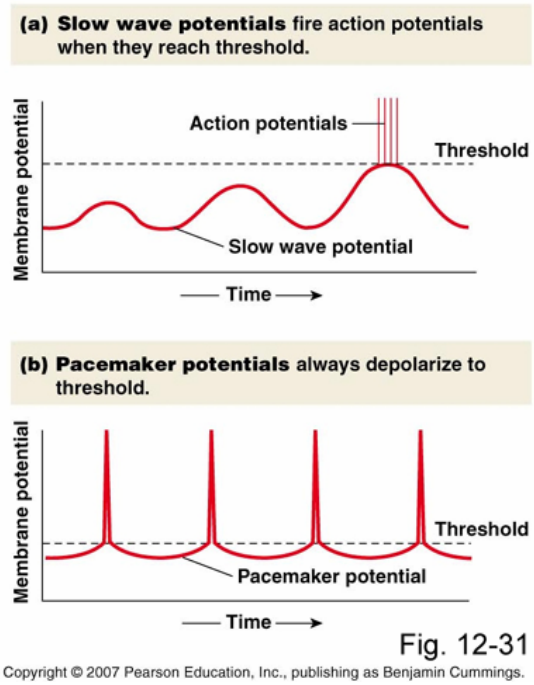


Fig. 12-31

Note – There is no RMP → membrane potential is variable (fluctuates around – 50 mV)

- (2) Multiunit smooth muscle
  - Found in eye (iris and ciliary body), large blood vessels, small airways, hair follicles
  - These smooth muscle cells are not linked together by gap junctions

- Require ANS-mediated neuronal generation of an action potential for contraction
- Activity of contraction mediated by (i) ANS and (ii) Hormones/paracrine agents

(III) Excitation-contraction coupling and relaxation of smooth muscle:

- Contraction:
  - (i) Influx of extracellular Ca^{2+} via voltage-gated or ligand-gated membrane channels → causes ↑ IC [Ca^{2+}]
  - (ii) Calmodulin (an IC Ca^{2+}-binding protein) binds up to 4xCa^{2+} → forms a Ca^{2+}-Calmodulin complex
  - (iii) This complex activates Myosin Light Chain Kinase (MLCK) → phosphorylates myosin light chain → activates myosin ATPase → cross-bridging/contraction
- Relaxation:
  - (i) Ca^{2+} removed by – (a) Ca^{2+} ATPase, and (b) Ca^{2+}/Na^{+} antiport
  - (ii) Myosin Light Chain Phosphatase → prevents myosin ATPase activation by dephosphorylating the myosin light chain

(IV) Smooth muscle has distinct functional qualities:

- (1) Latch-bridging – Despite a ↓ in cytoplasmic [Ca^{2+}] and dephosphorylation of myosin light chain, there can be sustained smooth muscle contraction with minimal energy expenditure (O_2 and ATP) as myosin head continues to remain attached to actin
- (2) Plasticity – There is a highly variable tension that develops in smooth muscle at a given length → when stretched, it tends to contract BUT the tension that develops changes with time despite a constant length

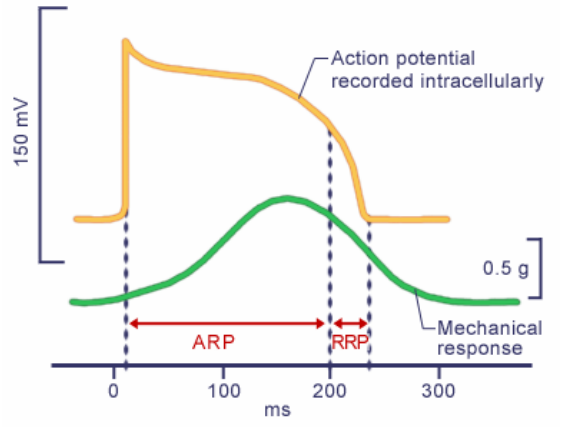
Cardiac Muscle:

(I) Anatomy of cardiac muscle:
- Single nucleus
- Mitochondria-rich (and as a result, has a VERY high O_2 demand)
- Striated (pattern less ordered cf. skeletal muscle)
- Possess "Intercalated Disks" – Specialised junctions b/t cardiac cells that permit ALL cardiac muscle contract together in a coordinated manner → these disks contain:
    - (a) Desmosomes – Connections b/t cells that give mechanical stability
    - (b) Gap junctions – Low resistance junctions that allows direct movement of ions from one cell to another → facilitates spread of APs between cells

(II) Type of cardiac muscle:
- (1) Contractile cells
    - Found in the atria and ventricles → contracts to generate stroke volume
    - Depolarise by spread of AP from surrounding pacemakers cells and contractile cells via "gap junctions" → influenced by ANS and hormonal control
- (2) Autorhythmic cells
    - Found in conducting pathway (SAN, AVN, bundle of His, Purkinje fibres) → SAN generates pacemaker potential and conducts it throughout the heart
    - Spontaneously depolarise → influenced by ANS and hormonal control

(III) Excitation-contraction coupling and relaxation of cardiac muscle:
- Excitation-contraction coupling occurs via "Calcium-induced calcium release":
    - (i) AP arrives at cardiac muscle cell → spreads into interior of cell by T-tubules
    - (ii) This opens up VG L-type Ca^{2+} channels (as part of "plateau" phase of cardiac muscle AP) → causes influx of extracellular Ca^{2+}
    - (iii) This triggers a substantial amount of Ca^{2+} release from the SR → SR Ca^{2+} then binds Troponin → contraction
    - Note – This mechanism allows for "Graded contractions" → where the degree of force depends on amount of Ca^{2+} entry!
- Relaxation occurs as Ca^{2+} is pumped back by:
    - (i) Ca^{2+} ATPase into the SR
    - (ii) Na^+/Ca^{2+} exchange – Needs Na^+/K^+ ATPase ↓ IC $[Na^+]$ to facilitate extrusion of Ca^{2+}

(IV) Repeated stimulation of cardiac muscle:
- It is IMPOSSIBLE to summate contractions or tetanise cardiac muscle because:
    - (1) Duration of cardiac muscle contractile response (300 msec) is as long as the duration of the cardiac muscle AP (250 msec)
    - (2) Cardiac muscle AP has a LONG refractory period – (i) Absolute refractory period → first 200 msec (up to phase 3) as "fast" Na^+ channels are still open, and (ii) Relative refractory period → last 50 ms (after phase 3) as "fast" Na^+ channels reset



<u>Differences between skeletal, smooth and cardiac muscle</u>:

Characteristics		Skeletal muscle	Smooth muscle	Cardiac muscle
Type of cells		(i) Extrafusal fibre (type I, IIa, IIb muscle) (ii) Intrafusal fibre (muscle spindle)	(i) Visceral (single unit) SM (GIT, ureter, vessel) (ii) Multiunit SM (eye, airway, vessel, follicle)	(i) Contractile cell (atria/ventricular) (ii) Autorhythmic cell (pacemaker)
Location		Attached to skeletal framework	Hollow viscera (GIT, ureters, uterus), blood vessels, bronchioles, eyes, hair follicles, Etc.	Heart
Function		Movement of body in relation to the external environment	Varies with structure (Eg. movement of contents within hollow organs, control of vascular and bronchial tone and resistance, piloerection, uterine tone, Etc.)	Generating cardiac output (HR and SV) for the systemic and pulmonary circulation
Structure	Striation	Present	Absent	Present
	Nuclei	Multi-nucleated	Single nucleus	Single nucleus
	Intercellular connections	Absence of gap junctions/desmosomes	Possess gap junctions	Possess intercalated discs (gap junctions and desmosomes)
	Contractile element	- Actin-myosin filaments arranged in interdigitating manner → striated - Troponin-tropomyosin complex on actin - Z-lines fix actin to membrane	- Actin-myosin filaments diagonally arranged → lack striation - Tropomyosin on actin only - Dense bodies fix actin to membrane	- Actin-myosin filaments arranged in interdigitating manner → striated - Troponin-tropomyosin complex on actin - Z-lines fix actin to membrane
	Sarcotubular system	Well-developed	Poorly developed	Poorly developed
Energy source		- Rich in mitochondria → ↑ [O] phosphorylation of CHO and fats - Creatinine phosphate energy stores	- Very few mitochondria → ↑ substrate phosphorylation (glycolysis) of CHO - No creatinine phosphate energy stores	- Rich in mitochondria → ↑ [O] phosphorylation of fats - Creatinine phosphate energy stores
Innervation and control	Stimulation of contraction	Nervous innervation by somatic NS	- Spontaneous (autorhythmic) → visceral SM - Nervous innervation by ANS → multiunit SM	- Spontaneous (autorhythmic) → pacemaker cell - Local AP spread via gap junctions → contractile cell
	Generation of muscle AP	"Motor unit" – Motor nerve AP → releases ACh → stimulates MEP and depolarises muscle fibres associated with the motor nerve	Local spread of AP between cells via gap junctions	Local spread of AP between cells via gap junctions
	Control of contraction	Nervous innervation by somatic NS	Influenced by SM stretch, ANS, hormonal and paracrine factors	Influenced by ANS, and hormonal factors
	Voluntary / involuntary	Voluntary	Involuntary	Involuntary
	RMP	Stable (-70 to -90 mV)	Wandering (- 50 mV)	- Wandering → pacemaker cell - Stable (- 90 mV) → contractile cell
	Muscle AP duration	Short (1-3 msec)	Short	Long (150-250 msec)
Contractile function	Sliding-filament	Present – Repeated cycles of actin-myosin head cross-bridge attachment, pulling of actin	Present (as skeletal muscle)	Present (as skeletal muscle)

	theory	fibres by myosin head towards centre of sarcomere, and detachment of actin-myosin head cross bridging		
	Excitation contraction coupling	- Muscle AP conducted via T-tubules → triggers DHP Ca^{2+} → charge movement triggers Ryanodine receptor → Ca^{2+} release from SR - Ca^{2+} binds to troponin → shifts tropomyosin → exposes actin and removes inhibition of myosin ATPase → permits actin-myosin cross-bridging	- Muscle AP opens voltage- or ligand-gated Ca^{2+} channels → influx of extracellular Ca^{2+} - Ca^{2+} binds to Calmodulin → forms complex that activates MLCK → phosphorylates myosin light chain → activates myosin ATPase → permits actin-myosin cross-bridging - Lacks troponin	- Muscle AP opens VG L-type Ca^{2+} channels → influx of extracellular Ca^{2+} → triggers Ca^{2+} release from SR (Calcium-induced calcium release) - Ca^{2+} binds to troponin → shifts tropomyosin → exposes actin and removes inhibition of myosin ATPase → permits actin-myosin cross-bridging
	Duration of contraction	Brief (7.5 to 100 msec)	Sustained	Moderate (300 msec)
	Muscle tone	None	Present	None
	Gradation of muscle strength	(i) Twitch summation → tetany (ii) Recruitment of motor units	Varying cytosolic [Ca^{2+}] by ANS and hormonal factors, and stretching	- (i) Varying cytosolic [Ca^{2+}] by ANS and hormonal factors, and (ii) Varying resting length of fibre (depends on filling volume) - CANNOT undergo tetany
	Length-tension relationship	Present	Absent	Present
	Others	Uses muscle spindles and golgi tendon organs to regulate contractions	- Latch-bridging – Despite a ↓ in cytoplasmic [Ca^{2+}] and dephosphorylation of myosin light chain, there can be sustained smooth muscle contraction with minimal energy expenditure as myosin head continues to remain attached to actin - Plasticity – There is a highly variable tension that develops in smooth muscle at a given length → when stretched, it tends to contract BUT the tension that develops changes with time despite a constant length	Lacks features spindles, golgi tendon organs, latch bridging and plasticity