Physiology week 2 – Excitable Tissue

Resting membrane potential

–70mV

Develops by:

1. NaK ATPase pumps Na out and K in
2. membrane permeable to Na and K which leak down [ ] gradients
3. membrane 100x more permeable to K than Na so much more K lost from cell
    a. net result → more + leaves cell → net – charge inside
4. other small factors
    a. Na pump cause 3Na out:2K in → loss + charge
    b. large intracellular proteins/organic phosphates can't diffuse → results in redistribution of small diffusible ions across membrane ie Gibbs-Donnan effect

Ions that contribute most to RMP have: high [ ], large [ ] gradient, large membrane perm (ie K)

So RMP similar to equilibrium potential for K ~ -90mV

Nerve AP generation

Stimulus may be electrical, chemical or mechanical.

This is maintained by Na/K ATPase pump.

Resting membrane potential -70mV- latent period

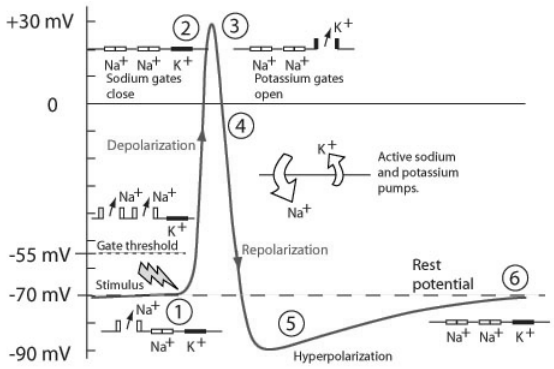
Gradual 15mV depolarization until firing threshold -55mV

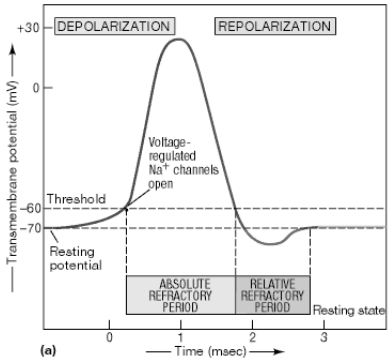
Overshoot (rapid depolarization) due to opening of voltage-gated Na channels to +35mV

Repolarization (Na channels inactivated, voltage gated K channels open, K leave cell) - rapid then slowly

After-hyperpolarization (overshoots) due to voltage gated K channels (which have slower opening and more prolonged than Na) beyond -70mV

Slow return of K channels to closed state/K diffuses away – back to RMP





Ion fluxes during AP

Fast Na influx (depolarization)

Na channels rapidly close (inactivated state)

Voltage-gated K channels open

Slow K efflux (repolarization)

Decr extracellular Ca decreases Na and K conductance required for AP

Refractory Period

Period where nerve is refractory to stimulation

Absolute - From firing time to one third completion of repolarisation - Na channels open or recovering

Relative - From one third completion of repolarisation to start of hyperpolarisation - K channels open

Electrogenesis of AP

Nerve cell membrane is polarized at rest with positive charges lined up along outside of the cell.

During AP, polarity briefly reversed.

Positive charges ahead and behind flow into the area of depolarization (current sink).

By drawing positive charges this makes the surrounding membrane approach firing level.

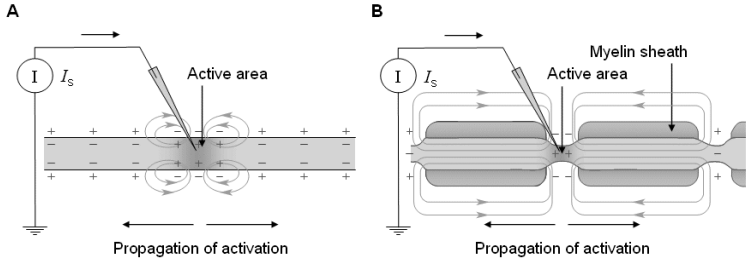
The area behind the AP is refractory therefore the AP moves forward

Where are ion channels distributed in myelinated neurons?

1. voltage-gated Na channels concentrated in node of Ranvier and initial segment
2. Na channels flanked by K channels

Saltatory conduction

In myelinated nerves - myelin effective insulator → depolarization jumps from one node of Ranvier to next
50x faster than unmyelinated



Factors that affect conduction in nerve cell

1. Myelinated vs demyelinated
2. Saltatory vs non-saltatory
3. Size
4. Direction of conduction

Ionic basis of excitation and conduction

Slight ↓ RMP → ↑ K out and Cl in → restores RMP

If depolarization reaches threshold potential → voltage-gated Na channels activated → influx Na along [ ] gradient → membrane potential can be 30-35mV (inside +) → repolarized by:

- Na channels rapidly closed/inactivated
- Direction electrical gradient Na reversed during overshoot (limits Na influx)
- Opening voltage-gated K channels → K out → slowly K channels closed (hyperpol)

IF:

- ↓ external Na → ↓ size AP, little effect on RMP (Na perm low)
- ↑ external K → ↓ RMP
- ↓ extracellular Ca → ↑ excitability by ↓ amount depol necessary to initiate changes in Na/K conductance
- ↑ extracellular Ca → stabilizes membrane/↓ excitability

Nerve fibre types

Erlanger and Gasser classification			
Type	**Function**	**Diameter**	**Velocity**
A alpha	Proprioception, somatic motor	15um	100m/s
A beta	Touch, pressure	10um	50m/s
A gamma	Motor to muscle spindles	5um	25m/s
A delta	***Pain***, temperature	3um	20m/s
B	Preganglionic autonomic	1um	1m/s
C (unmyelinated)	***Pain***, temp, itch, reflexes, postganglionic sympathetic	1um	1m/s

Greater diameter = greater speed

A + B myelinated

C unmyelinated

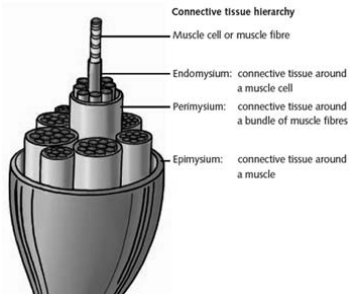
Susceptibility to: (most → least)

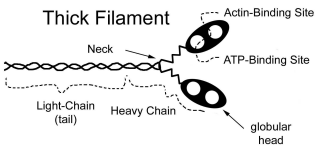
Hypoxia	B>A>C
Pressure	A>B>C
LAs	C>B>A

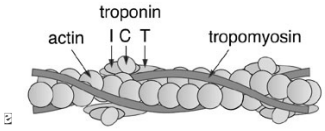
Muscle Morphology

Structure

Each muscle fibre a single cell surrounded by sarcolemma.
Fibre made up of myofibrils, myofibrils made of filaments, filaments made of contractile proteins.
Thin filaments - actin (forms 2 chains in long double helix), tropomyosin, troponin.
Thick filaments - myosin (2 globular heads, long tail).



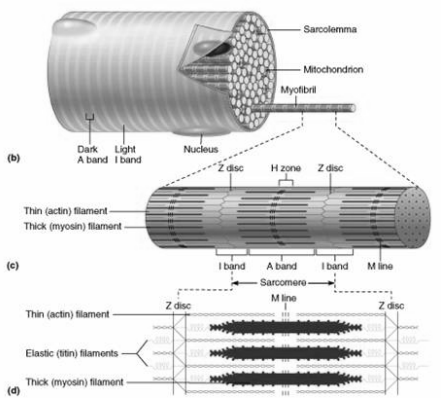




Striations

Caused by arrangement thick and thin filaments.
Area between two Z lines is sarcomere.
Thick filaments form the A band.
Thin filaments form the I band.

T tubules enter between A and I.
Each T tubule has a cistern on either side.
M lines are at centers of the A bands.



Electrical characteristics of skeletal muscle

RMP -90mV AP 2-4 ms

Motor unit

Single anterior horn alpha-motor neuron, its axon, all muscle fibres innervated - functional unit of contraction
Smaller number of fibres = finer movements

Mechanism of skeletal muscle contraction

Sliding filament hypothesis - myosin head flexes (ATP hydrolysis), shortens, actin/myosin crosslink

Excitation-contraction coupling

- Sequence of events from muscle excitation to muscle contraction

Discharge of motor neuron
Release of ACh from motor endplate
ACh binds postsynaptic nicotinic ACh receptors
Increased Na/K conductance in end plate membrane.
Generation EPP (end plate potential)
When threshold reached (50-55mV) AP occurs in muscle fibres.
Inward spread of AP via T tubules
Activates voltage-gated L-type Ca channels (dihydropyridine receptors)
Ca enters cell and Ca released from SR (ryanodine receptors)
Ca bind trop C, weaken trop I bindings, causing tropomyosin to move laterally, expose actin/myosin binding sites.
Myosin head binds actin chain. Thin filaments slide on thick
ATP enters ATP binding site, is hydrolysed, causes power stroke, shortens sarcomere by 10nm

Relaxation:

Ca pumped back into the SR by a Ca Mg pump that requires ATP.
Release of Ca from trop C.
Cessation of interaction between actin and myosin

Type of contraction

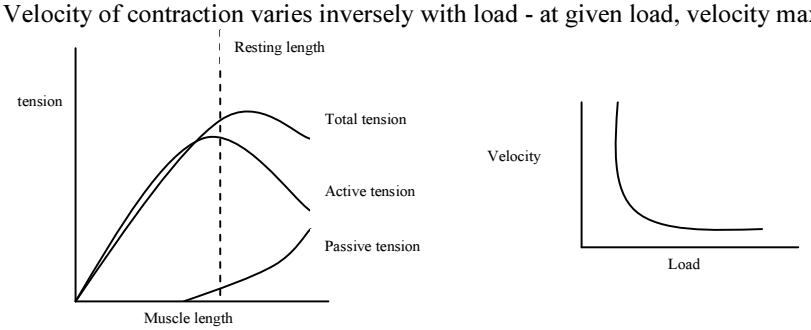
Isometric - doesn't shorten; Work = force x distance → isometric goes no distance so less work
Isotonic - contraction against constant load, muscle shortens

Summation of contractions

Electrical response of a muscle fibre to repeated stimulation.
Contractile mechanism has no refractory period so repeat stimulation before relaxation adds up - summation
Rapid repeat stimulation → fuse into one continuous contraction - Tetanic contraction → tetanus
Complete (no relaxation between stimuli, tension developed ~4x that of individual twitch contraction)
Incomplete (periods of incomplete relaxation between summated stimuli)
Treppe effect (staircase phenomenon) → if series max stimuli delivered at frequency just below tetanizing frequency there is increase in tension until plateau reached – due incr Ca availability

Relation between muscle length, tension and velocity of contraction

Total tension (tension muscle develops when stimulated to contract isometrically)
Active tension
Passive tension (tension exerted by unstimulated muscles)
Total and passive tension vary with length
Active = total – passive at any given length
Resting length = length of muscle at which active tension maximal
Tension develops in proportion to number of actin-myosin cross links
When muscle stretched overlap of actin-myosin decreases so number of cross links decreases
When muscle shortened distance thin filaments can move is decreased
Velocity of contraction varies inversely with load - at given load, velocity maximal at resting length



Muscle fibre types

	Type 1	**Type 2**
	slow, oxidative, red	fast, glycolytic, white
Function	maintain posture eg. mm back	fine movements eg extraocular, hand mm
Twitch duration	long	short
Ca pump capacity SR	moderate	high
Diameter	moderate	large
Glycolytic capacity	moderate	high
Oxidative capacity	high	low
	slow myosin ATPase rate, fatigue resistant red due to high content myoglobin	fast myosin ATPase rate, fatiguable

Can change muscle function by activity, innervation or hormones.
Motor unit only contains one type of fibre

Energy sources

Ultimate source of energy: $ATP + H_2O \rightarrow ADP + H_3PO_4 + 7.3$ Kcal
Sources of ATP:
Phophorylcreatine
Energy rich phosphate compound used to supply energy for short periods.
Hydrolysed to creatine and phosphate with release of ATP.
Glucose + 2ATP → 2lactic acid + 4ATP
Glucose + 2ATP → $6CO_2 + 6H_2O$ + 4ATP (citric acid cycle)
FFAs → $CO_2 + H_2O$ + ATP

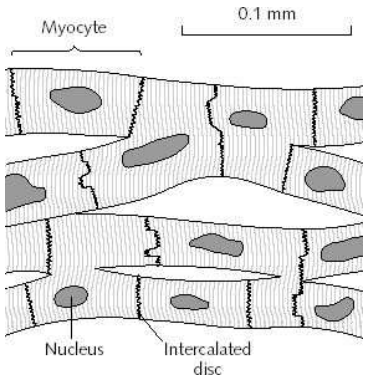
At rest and during light exercise muscles utilize FFAs.
More intense exercise requires carbohydrates
Glucose may come from blood stream or from breakdown of glycogen stores.

Oxygen debt

If oxygen is insufficient, pyruvate reduced to lactate.
After exercise, extra oxygen required to remove lactate, replenish ATP and phosphorycreatine stores
Athletes can increase O2 consumption and utilize ffa's more effectively → smaller debt

Cardiac muscle

Characterized by intercalated discs and gap jxns
Long, cylindrical cells with 1-2 nuclei centrally
Striations with Z lines
Fibres branch and interdigitate
Intercalated discs (where end of one fibre abuts another), provide strong union between fibres, have very low resistance and thus cardiac muscle termed a syncytium
Cell membranes along discs fuse form gap jxns
T system located at Z lines
Contains: myosin, actin, tropomyosin, troponin, titin, dystrophin



Skeletal vs cardiac muscle

	Skeletal	**Cardiac**
Depolarization	Nerve AP to each muscle cell, then to muscle endplate	Spread by specialized muscle cells in conducting system then cell to cell
Role of voltage-gated Ca channels (dihydropyridine receptor)	Acts as trigger to release of Ca from SR. Only small amounts extracell. Ca required	Similar: Ca enters cell, triggers release Ca from SR
Depolarization duration	Short	Long (150-250msec)
Contraction duration	Depends on fibre type, 7.5-10ms	At normal HR ~300ms in ventr
Muscle spindles	Present	Not present
Control	Voluntary	Involuntary
Plateau	Only fast Na channels open	Fast Na and slow Ca channels open

Cardiac muscle action potential

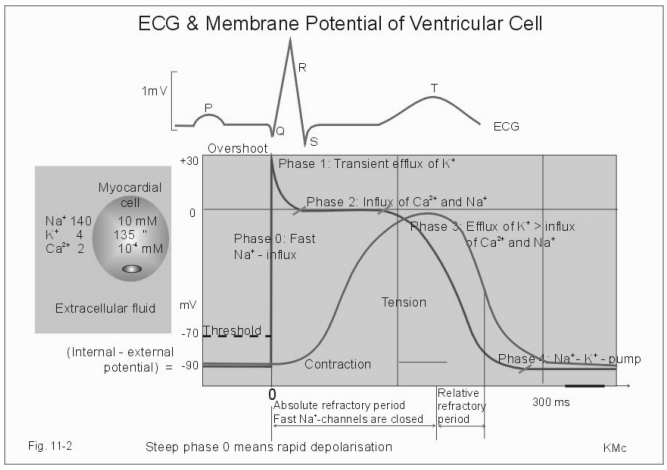
-90mV to +20mV
200ms duration
Characterized by intercalated discs and gap junctions.
Intercalated discs have very low resistance and thus cardiac muscle is termed a syncytium.

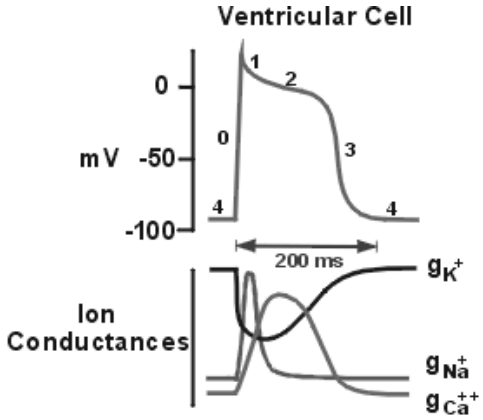
Phase 0 - Rapid depolarisation and overshoot to +20mV - opening voltage gated Na channels → rapid Na influx **(Na in)**
Phase 1 - Initial rapid repolarisation to 0mV - closure of voltage gated sodium channels
Phase 2 - Prolonged plateau - slow, prolonged opening voltage gated Ca channels → calcium influx **(Ca in)**
Sodium also flows through these channels
Reduced permeability to potassium during this phase
Phase 3 - Late rapid repolarisation - closure of voltage gated Ca channels
K efflux via various K channels (inward rectifying, delayed rectifying and transient outward K channels) **(K out)**
Phase 4 – RMP, -90mV

Relations to ECG:

- upstroke on QRS
- plateau occupies QT interval
- repolarisation at T wave

Tetany does not occur in cardiac muscle because muscle is still contracting in relative refractory period and beyond the duration of AP. (1.5x as long as AP)



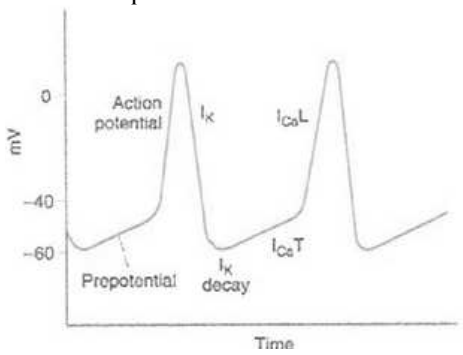


(rapid Na in, K out, slow Ca in)

Cardiac pacemaker AP

Resting membrane potential –60mV

1. pre-potential initially due to decr in efflux K, then completed by influx Ca via T channels
    a. automaticity due to rising prepotential (leakage of K/Ca)
2. AP due to influx Ca via L channels
3. repolarisation due to efflux K
4. no plateau



How do autonomic (PSNS/SNS) factors alter the slope of the prepotential?

NA:

1 Noradrenaline from sympathetic endings binds beta 1 receptor raises intracellular cAMP

2 Facilitates opening of L channels

3 Increased Ca2+ influx

4 Increased heart rate and increased slope of prepotential

ACh:

1 binds M2 muscarinic receptors and G protein, decreasing cAMP

2 results in opening K+ channels

3 decreased slope of prepotential and decrease firing rate

Compare cardiac myocyte AP and pacemaker AP

1. Na fast vs Ca dependent
2. automaticity due to rising prepotential (leakage of K/Ca)
3. plateau phase due to Ca
4. > resting potentials

Compare APs in peripheral myelinated axon/skeletal muscle and cardiac muscle

Peripheral nerve much shorter (1-2msec cf 5-10msec)

Absence plateau

Absence of pre-potential

Determinants of force of contraction of cardiac muscle

1. muscle fibre length (Starling's curve)
2. catecholamines
    a. via beta1 adrenergic receptors and cAMP
    b. cAMP activates protein kinase A → phosphorylation of voltage dependent Ca channels → spend more time in open state
    c. cAMP also increases active transport Ca into SR → accel relaxation, shortening systole
3. digitalis
    a. increased contraction by inhibition of NaK ATPase
    b. increases intracellular Na → decreased Na gradient across membrane → decrease Na influx and Ca efflux → increase intracellular Ca

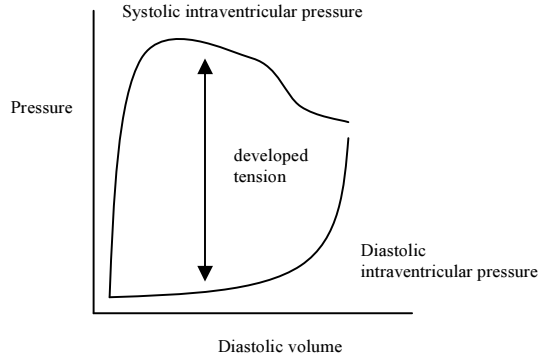
Length-tension

Resting length = length at which tension developed on stimulation is maximal

Initial length of fibres determined by: Degree of diastolic filling - pressure developed in ventricles proportional to total tension - **Starling's law of the heart**

Therefore increase diastolic volume → increase tension until reaches maximum = ascending limb of Starling curve

Then tension tends to decrease (desc limb) → due to beginning disruption of myocardial fibres



<u>Smooth muscle</u>

Structure

No cross striations, Irregular actin/myosin chains, No troponin, Poorly differentiated SR, Few mitochondria
Dense bodies in cytoplasm, Single nucleus, Largely dependent on glycolysis

Types

Visceral

In large sheets, primary in walls of hollow viscera (intestine, uterus, ureters)
Low resistance bridges between individual muscle cells incl gap junctions
Function in syncytial fashion

Multi-unit

Made up of individual units without interconnecting bridges, nonsyncytial
Contractions do not spread widely – contracts more discrete, fine, localized
Sensitive to circulation chemicals or nerves
NA → repeated firing of muscle after single stimulus rather than single AP
Irregular tetanus rather than single twitch
Iris → fine, graded contractions
Not under voluntary control, but functionally similar to skeletal muscle

Contraction

Binding of Ach to muscarinic receptors
Increased Ca influx to cell via voltage-gated Ca channels
Calcium binds calmodulin and activates MLCK (myosin light chain kinase)
MLCK catalyses phosphorylation of myosin
Increased myosin ATPase activity
Actin slides on myosin → contraction
Dephosphorylation of MLCK
Relaxation or sustained contraction due to latch bridge and other mechanisms

Compare smooth muscle to skeletal muscle contraction

	Skeletal muscle	**Smooth muscle**
Major source of Ca	SR	ECF via voltage gated Ca chann
Ca binds to	Troponin C	Calmodulin
Fxning of this binding by Ca	Removes inhibition of trop I & results in exposure myosin binding sites	Ca – calmodulin activates MLCK
Activation of myosin ATPase	Doesn't require phosphorylation	Requires phosphorylation, catalysed by active MLCK
Contraction	Brief	Long
Control	Voluntary	Involuntary
Stimulation of contraction	Nervous innervation necessary to initiate	Often spontaneous without nervous stim → sensitive to circulating chemicals
RMP	Stable	No RMP as potential wanders

Compare cardiac and smooth muscle contraction

Cardiac muscle	**Smooth muscle**
Responses PHASIC → contraction alternates with relaxation	Contraction often tonic due to latch bridge mechanism
Increased intracellular cAMP → increased force of contraction	cAMP → relaxation of vascular smooth muscle as it inhibits phosphorylation of MLCK

Electrical and mechanical activity of smooth muscle

Characterized by:

Instability of membrane potential
Shows continuous, irregular contractions independent of nerve supply
Maintained state of partial contraction = tone
Membrane potential has no true resting value
Low when tissue active, High when inhibited, In relative quiescence -50mV
Superimposed on membrane potential are waves - slow, sine wave-like fluctuations
+ pacemaker potentials → generated in multiple foci that shift
Excitation – contraction coupling in smooth muscle a slow process

Factors that stimulate visceral smooth muscle

1. Contracts when stretched in absence of extrinsic innervation - stretch → decr membrane potential → inc freq spikes and incr tone
2. Nadr and Adr - membrane potential larger, spikes decrease in frequency, muscle relaxes
3. β-adrenergic receptors - decreased muscle tension, mediated by cAMP, prob due to incr intracellular binding Ca
4. α-action - inhibition contraction due to increased Ca efflux from cell
5. Ach - membrane potential decr, spikes more frequent, muscle more active with incr tonic tension/no. rhythmic contractions - mediated by phospholipase C and IP3 → incr intracellular Ca
6. cold

Function of nerve supply to smooth muscle

1. spontaneous activity occurs in absence of nerve stimulation
2. sensitivity to chemicals from nerves locally of via circulation
    a. dual supply from PSNS and SNS
    b. function of nerve supply is not to initiate activity in muscle but to modify it

Relationship between length and tension in smooth muscle

Characteristic of smooth muscle: variability of tension it exerts @ any given length
→ if stretch smooth muscle, initial increase in tension but then tension gradually decreases
→ referred to as plasticity of smooth muscle
eg tension exerted by smooth muscle walls of bladder → initially little increase in tension as volume increased due to plasticity of bladder walls, but point reached @ which bladder contracts forcefully